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Orchid Stomata—Structure, Differentiation, Function, and Phylogeny*

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Introduction

In this review of orchid stomata it is a pleasure to include a number of little-known studies. These include several doctoral dissertations and works in languages other than English. My literature search brought much more information to light than I anticipated, and revealed a surprising variety in stomatal structure, function, and development in orchids. The survey of the available literature also demonstrated the shortcomings of our present knowledge. Readers will easily see which groups of orchids are the most popular subjects for study and which have received virtually no attention at all. They will also note that ultrastructural and physiological studies of stomata rarely employ orchid material. There is, of course, no reason to presume that our general knowledge of stomata in these other plants does not apply to orchid stomata. On the other hand, only the few studies that do involve orchids have been considered relevant for this review. For these reasons, a general comparison of orchid stomata with those of other plant groups seems premature.

The literature treated here covers a span of 150 years with considerable concentration on studies that are about 100 years old. Naturally, scientific traditions and methods have changed markedly during this period, and in comparing the old reports with more recent ones it is at times difficult to ascertain whether they really deal with the same kind of observations. Orchid nomenclature has also changed considerably. This development has made it necessary to cite all Latin orchid names with their authorities and often to "translate" them into modern names. In the latter case the name used in the source in question will appear in parentheses.

With our insufficient knowledge of intraspecific variation and the possibility of misidentified material in mind, it is hard to select between conflicting reports. I find it even more risky to reject published results when they are at variance only with casual or unpublished observations by myself or others. There are some observations, however, that seem so unusual that it would be best to have them confirmed independently; these observations will be pointed out in the text. I have reservations about a few sources both old and recent. I have little confidence in developmental studies that are based on young tissues but do not present mitotic figures to support the interpretations. Developmental conclusions drawn solely on the basis of cell configurations in the mature tissue I consider even less reliable.

The descriptions in this chapter are adapted to the terminology of Rasmussen (1981b) and Wilkinson (1980). In the glossary, special attention is given to the difficult terminology of stomatal development. As for the morphological terminology of mature stomata, terms such as "anisocytic" or "paracytic" are simply used descriptively for the mature stomatal complex, with no implications as to the origin of the cell involved. The orchid classification used here is in accordance with Dressler (1981).

Surface Structure

The first study of general leaf anatomy and stomatal characters in orchids is that by Möbius (1887) treating 193 species in 95 genera. Other comprehensive surveys are those by Solereder and Meyer (1930), which in addition to a review of previous literature includes their own data for 73 species, and Lavarack (1971) who observed leaf characters in about 150 species. Solereder and Meyer (1930) presented useful lists of stomatal sizes (p. 104), stomatal structure (pp. 104–105), and occurrence of subsidiary cells (p. 105).

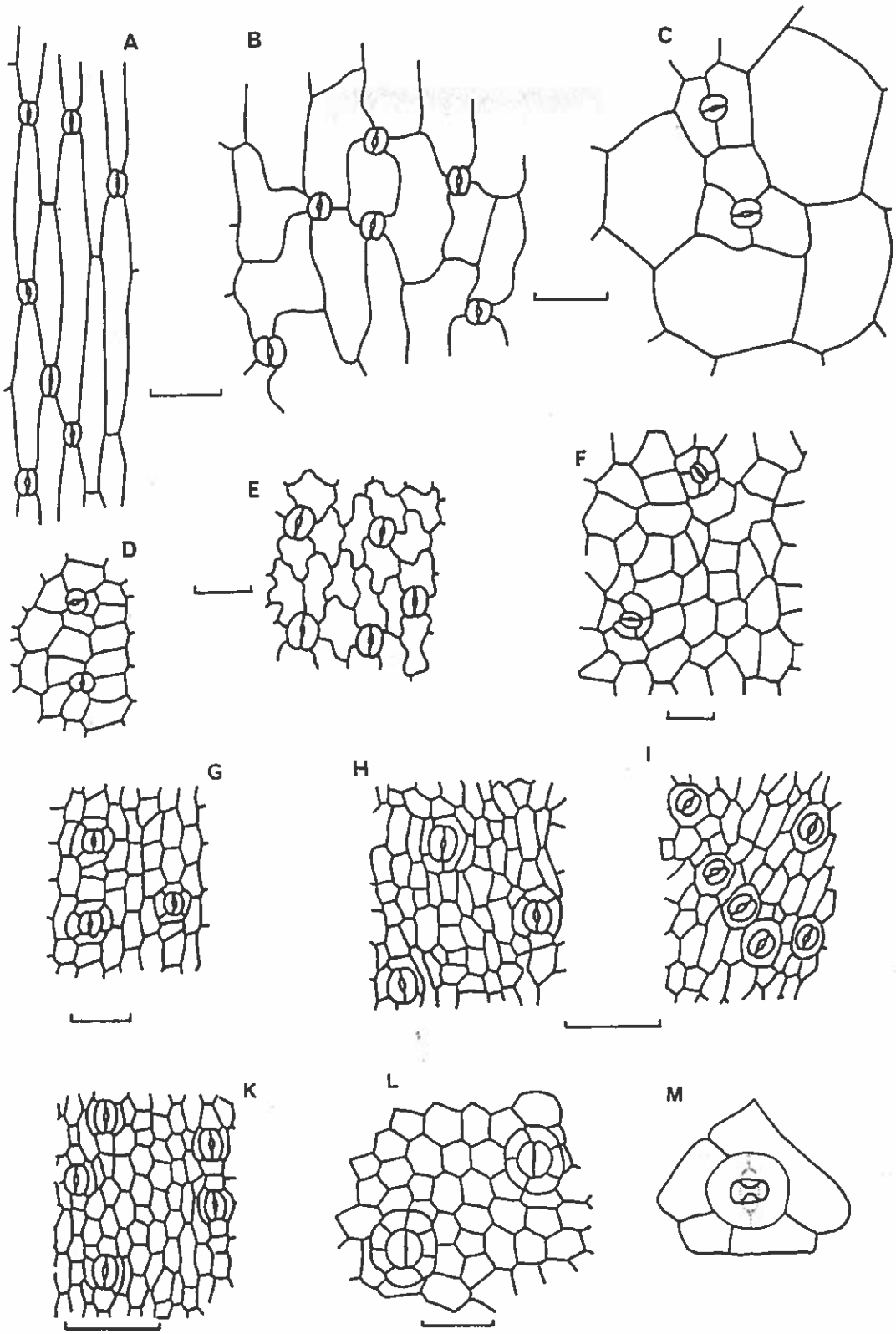
Generally, guard cells are oriented parallel to the long axis of the leaf (Siebe, 1903; Solereder and Meyer, 1930; and many others). Ziegenspeck (1944b) published a list of broad-leaved European orchids in which the number of deviations from longitudinal orientation of stomata were compared with the venation. According to this list, the longitudinal orientation prevails even when venation is curved. One clear-cut example to the contrary is *Goodyera repens* (L.) R. Br., in which the stomata are scattered. Cyge (1930) reported that in species with broad leaves and sinuous anticlinal epidermal cell walls the stomata are not all longitudinally oriented. Even under such conditions, however, a general longitudinal orientation is often maintained as, for instance, in *Listera cordata* (L.) R. Br. (Fig. 4-1-E; Raunkiær, 1895–99).

Longitudinal orientation of stomata is also observed in the rare cases where stem stomata have been found (Siebe, 1903; Nayar et al., 1976).

Guard Cells

Individual guard cells are reniform (kidney-shaped) and the outline of the pairs is elliptical (Fig. 4-1). Sprenger (1904) and Solereder and Meyer (1930), however, noted stomata with almost circular outlines in species of *Bulbophyllum*, including *Cirrhopetalum* (Fig. 4-1-M; Sprenger, 1904). Stomata that are broader than they are long were reported in *Pholidota imbricata* Lindl., *Sobralia macrantha* Lindl., and *Vanda teres* Lindl. (Solereder and Meyer, 1930). Peculiar angularly shaped guard cells were described in *Apostasia* (*Adactylus*, Siebe, 1903). Their polar ends were flat rather than rounded. Unfortunately, these cells were not illustrated.

Figure 4-1. Surface views of stomatal complexes in leaves of various orchids. A. *Diuris punctata* Sm., anomocytic stomata. B. *Cryptostylis subulata* (Labill.) Rchb. f., anomocytic stomata. C. *Zeuxine oblonga* Rog. & C. T. White, stomata probably best characterized as diacytic; anisocytic stomata according to Lavarack (1971). D. *Hammarbya paludosa* (L.) Kuntze and E. *Listera cordata* (L.) R. Br., anomocytic stomata. F. *Nervilia purpurea* (Hoyata) Schltr. and G. *Coelogyne occultata* Hook. f., tetracytic stomata. H. *Pholidota articulata* Lindl. var. *griffithii* K. & P., tetracytic stomata, which at a later stage (I) develop into floating stomata. K. *Vanda cristata* Lindl., paracytic stomata. L. *Dresslerella hispida* L. O. Wms., cyclocytic stomata. M. Detail of stoma from *Bulbophyllum* sp. showing the oblong opening in the cuticular ledges (bold outline) oriented at right angles to the stomatal pore (stippled). Scales: A, B, C, D, E, F, G, L = 0.10 mm; H, I = 0.25 mm; K = 0.50 mm, no scale indicated on original of M. (Sources: A–C, Lavarack, 1971; D–E, Raunkiær, 1895–99; F, Namba et al., 1981, courtesy of *Shoyakugaku Zasshi;* G, Banerjee and Rao, 1978, courtesy of *Current Science;* H–K, Singh and Singh, 1974, courtesy of *Current Science;* L, Pridgeon and Williams, 1979; M, Sprenger, 1904. All redrawn, L from photograph.)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Size of Stomata

The reported sizes of orchid guard cells vary greatly. According to Solereder and Meyer (1930, p. 104) a minimal length was noted in stomata of *Epidendrum equitans* Lindl. (20 μm), whereas a maximum of 75 μm was observed in *Trichoglottis solerederi* Kränzl. These authors noted a correlation of stomatal size with habitat—epiphytes generally have smaller stomata than terrestrial species. This finding is in agreement with the observation that stomatal sizes tend to increase from the top layer in a tropical forest toward the bottom when all plant groups are considered (Roth and de Bifano, 1979). The sizes of stomata in the epiphytic species of *Odontoglossum* and *Palumbina* (Ayensu and Williams, 1972) and *Myoxanthus* (Pridgeon and Stern, 1982) were all in the lower end of the size range. The same finding applies to species of *Vanilla* (Nayar et al., 1976), whereas in the terrestrial *Paphiopedilum* species (Rutter and Wilmer, 1979) the largest stomata, being 81 μm in length, even exceed the maximum described by Solereder and Meyer. In a sample consisting only of epiphytic species, Avadhani et al. (1982) noted an amazing range from 9 × 9 μm in *Phalaenopsis violacea* Teism. and Binn. to 110 × 70 μm in *Thrixspermum calceolus* (L.) Rchb. f. These measurements are of the outline of the outer ledges and therefore are not directly comparable to the dimensions listed earlier.

Cuticular Patterns

Cuticular patterns of the surface view of stomata may be quite confusing to the unexperienced observer. The outer cuticular ledges fuse to form anything from a ring to an almost complete cover over the stoma. Air trapped in the outer cavity may be the reason for the refractive effect observed by Faber (1904). The circumference of the cover usually corresponds to the outline of the guard cells, but the opening at the top does not always resemble the pore below. This opening may be a narrow slit, as in species of *Cymbidium* (David DuPuy, personal communication); a circular hole, such as in *Bifrenaria harrisoniae* Rchb. f., *Epidendrum ciliare* L., and *Vanda tricolor* Lindl. (Solereder and Meyer, 1930); or a bluntly cornered rectangle. Species of *Bulbophyllum* (Möbius, 1887; Sprenger, 1904), for instance *B. sessiliflorum* Kränzl., have the latter type of opening in which the longitudinal axis of the rectangle is oriented perpendicular to that of the pore. This appearance could be misinterpreted for transversely oriented stomata. A similar arrangement was noted by Hering (1900) in *Angraecum subulatum* Lindl. (*Listrostachys*) and by Solereder and Meyer (1930) in species of *Cattleya, Laelia,* and *Epidendrum.*

The outer cuticular cavity seen by Sprenger (1904) in some species of *Bulbophyllum* is elaborate. Two extensions bulge out from the polar ends of the cavity (Fig. 4-1-M), thereby segregating upper and lower portions of the cavity, which are connected by a narrow canal, the "double closure" (Doppelverschluss) of Solereder and Meyer (1930). Other species with protuberances from the polar ends of the outer cavity are *Papilionanthe subulata* (Koenig) Garay (*Aerides cylindricum* Lindl.) and *Laelia purpurata* Lindl. and Paxt. In the latter, the protuberances join, leaving a passage between the upper and lower portions of the cavity by only two, rarely three, small openings (Solereder and Meyer, 1930). Another peculiarity of the outer cavity is the presence of protrusions from the lateral sides (see later discussion of structure in cross section).

Around the cuticular elaborations of the guard cells, and separated from these by a ring-shaped slit, there is often a cuticular rim covering the subsidiary cells (Fig. 4-2-D; Zörnig, 1904, species of *Coelogyne;* Sprenger 1904, *Bulbophyllum fischeri* Seid. [*Cirrhopetalum gamblei* Hook.]). A similar development may be repeated in cells adjacent to the subsidiary cells (several examples in Sprenger, 1904). Rims running along the circumference of subsidiary cells may occur together with a cuticular bank on the subsidiary cells or alone (Zörnig, 1904, numerous species of *Coelogyne*). In many species of *Coelogyne,* a more extended pattern of ridges covers the surface of the whole stomatal complex (Zörnig, 1904).

Strasburger (1866–67) observed radiating (cuticular?) ridges on guard cells of *Pleurothallis pulchella* (HBK) Lindl. (*Stelis pulchella*), which in this respect were compared with those of *Equisetum.* In *Equisetum,* however, these ridges do in fact belong to two flanking and overarching subsidiary cells, according to Dayanandan and Kaufman (1973).

Guard Cell Walls

Plant cell walls contain a cellulose reinforcement, which is often characterized by a predominant orientation. The microfibrils constituting this reinforcement are observable in the electron microscope but their orientation can also be detected indirectly in the polarization microscope by means of their birefringence. By the latter method, Ziegenspeck (1936, 1944b) discovered that the microfibrils radiate from the stomatal pore in orchid guard cell (Fig. 4-4-A and B, species of Orchideae). This pattern later proved to be a feature of all kidney-shaped guard cells, and is believed to be responsible for the shaping of the guard cells and the development of a pore between them, as well as for the movements of the functioning stoma (Ziegenspeck, 1936; Aylor et al., 1973; see later discussion of function).

Uneven secondary thickenings of the guard cell walls are best described in transection (discussed later). A single interesting detail, which was observed in surface view and longitudinal section only, is the wall thickening in the polar ends of the guard cells in *Phragmipedium* species seen as a knot in each end of the stoma (Faber, 1904, tab. III, 46–47, tab. IV, 48; Solereder and Meyer, 1930). This thickening was not observed by Rosso (1966).

Contents and Fine Structure of Guard Cells

Oil droplets in guard cells have been observed in stomata of Coelogyninae (Zörnig, 1904) and *Paphiopedilum* (Faber, 1904; Rutter and Willmer, 1979). The survey by Ziegenspeck (1944a) of the occurrence of oil in guard cells indicated that oil is most often found in stomata of long-lasting and evergreen leaves. The orchid species included in that survey were all temperate, terrestrial species that were shown to lack oil.

Starch in guard cells was noted in *Corybas* (Groom, 1895) and in *Paphiopedilum* (Rutter and Willmer, 1979). In contrast, *Chamaeorchis alpina* (L.) L. C. Rich. and *Nigritella nigra* (L.) Rchb. f., as well as species of *Orchis* (including *Dactylorhiza*) and *Ophrys,* lack starch (Ziegenspeck, 1936).

Chlorophyll is generally present in guard cells, but ultrastructural studies and fluores-

cence microscopic tests demonstrated a lack of chlorophyll in stomata of species of *Paphiopedilum* (Nelson and Mayo, 1975; Rutter and Willmer, 1979). There are only a few positive reports on the presence of chloroplasts in orchid guard cells, such as Siebe (1903) in *Neuwiedia* and *Apostasia,* or on the absence of chlorophyll, such as MacDougal (1899) in the guard cells of the hemisaprophyte *Cephalanthera austinae* (A. Gray) Heller (*C. oregana* Rchb. f.). It is possible that the presence of chloroplasts was often taken for granted and no steps were taken to distinguish them from colorless plastids (leucoplasts) in the preparations used. When chloroplasts were specifically noted in the guard cells of *Pholidota pallida* Lindl. (Zörnig, 1904), it is possible that they were rare or absent in the other orchid species studied by Zörnig, or simply that they were most prominent in that particular species.

An ultrastructural study of *Paphiopedilum* stomata revealed quite remarkable features (Rutter and Willmer, 1979). Besides containing starch, the colorless plastids of the guard cells contain an elongated, fibrous body suggested to consist of stored protein. This type of plastid is considered to differ markedly from previously known types. The cytoplasm also contains numerous oil droplets.

How is the function of the stomata affected by the unusual structure and lack of chlorophyll (see discussion of function), and to what extent do these unusual features occur in other orchids? Lack of chlorophyll in functional guard cells has been demonstrated only in species of *Paphiopedilum.* On the other hand, this is the only orchid genus in which the ultrastructure of stomata has been studied so far.

Configuration of Subsidiary Cells

The stomatal meristemoid in monocotyledons is most often initially surrounded by four epidermal cells. This is due to the origin of the meristemoid as a small, apical segregate from an epidermal cell, and the fact that the epidermis consists of regular longitudinal cell files (Fig. 4-3-A; Strasburger, 1866–67). Irregularity of the second condition leads to higher or lower number of cells abutting the meristemoid.

A special case was described by Williams (1975) in *Ludisia discolor,* in which the meristemoid is formed by a curved wall that meets the apical anticlinal wall of the maternal epidermal cell twice. As a result, the lens-shaped meristemoid, and later the stoma, become surrounded by only two epidermal cells. This condition, which can be called diacytic, has been seen in *Zeuxine oblonga* R. S. Rogers and C. T. White (Fig. 4-1-C; Lavarack, 1971), *Ludisia discolor* (Ker-Gawl.) A. Rich., *Zeuxine strateumatica* (L.) Schltr., *Sarcoglottis acaulis* (J. E. Smith) Schltr. (*Spiranthes acaulis*), and *Prescottia stachyoides* (Sw.) Lindl. (Williams, 1975), as well as in *Beadlea elata* (Sw.) Small (*Spiranthes elata,* Dressler, 1981). Unfortunately, Lavarack termed this condition "anisocytic," and used this term in an unusually broad sense to includes paracytic and tetracytic conditions, as far as can be judged from his tables. Hence, apart from those cases for which he has shown illustrations, we cannot use Lavarack's studies to evaluate how frequently this interesting condition occurs. The anomocytic condition (Fig. 4-1-A, B, D, E) often occurs in orchids (Solereder and Meyer, 1930; Lavarack, 1971; Shah et al., 1975).

The tetracytic condition of orchid stomata (Fig. 4-1-F, G) is also very common (Sol-

ereder and Meyer, 1930). Except for *Pleione*, which was found to have anomocytic stomata, the genera of Coelogyninae show mostly tetracytic stomata (Zörnig, 1904). Tetracytic stomata were also found in *Bulbophyllum* (Möbius, 1887, tab. XXIII, 12; Sprenger, 1904); numerous Pleurothallidinae (Hünecke, 1904); *Paphiopedilum* and *Phragmipedium* (Rosso, 1966); *Odontoglossum* (Ayensu and Williams, 1972); *Vanda*, *Calanthe*, and *Rhynchostylis* (Singh and Singh, 1974); *Aerides* and *Phajus* (Shah et al., 1975); *Vanilla* (Nayar et al., 1976); *Myoxanthus* (Pridgeon and Stern, 1982); species of Cymbidieae and Maxillarieae (Williams, 1976); and numerous other species (Williams, 1979, table 1).

The tetracytic condition is dependent on a regular epidermal pattern, in which exactly four cells of approximately equal size abut the guard cell pair. Where, owing to an irregular epidermis, the condition of three, five, or more cells encircling the meristemoid is frequent, the general appearance is usually described as anisocytic (Fig. 4-1-F, H; Lavarack, 1971; Nayar et al., 1976; Banerjee and Rao, 1978; Namba et al., 1981).

A cyclocytic condition—that is, a ring of more than four subsidiary cells—was observed a few times in *Vanilla* (Benecke, 1892) and *Dresslerella* (Fig. 4-1-L; Pridgeon and Williams, 1979). A radial subdivision of four original subsidiary cells is supposed to be the origin of this condition (Pridgeon and Williams, 1979), but the ontogeny has not been described.

There are three reports of floating stomata in Orchidales. Singh and Singh (1974) observed floating stomata in a species of *Pholidota*. This condition is suggested to arise when anticlinal walls between subsidiary cells in a tetracytic configuration dissolve (Fig. 4-1-H, I; Singh and Singh, 1974). Besides *Pholidota*, this process was also noted to some degree in two species of *Coelogyne* (Singh and Singh, 1974), which were both described simply as tetracytic by Banerjee and Rao (1978). On the other hand, Banerjee and Rao (1978) observed floating stomata in two other species of *Coelogyne*. Singh (1981) confirmed the report of floating stomata in *Pholidota*.

Some orchid stomata are described as paracytic (Fig. 4-1-K). Examples are *Neuwiedia* and *Apostasia* (Siebe, 1903), *Aerides* (Shah et al., 1975), and *Vanda* (Singh and Singh, 1974). The numerous cases of two subsidiary cells observed by Williams (1979) in Oncidieae can also be described as paracytic.

Recognition of Subsidiary Cells

As outlined in the preceding section, the number of subsidiary cells in orchids varies from zero (anomocytic stomata) to six or seven (cyclocytic stomata). Occasionally, several contradicting reports exist for the same orchid group. For instance, in *Paphiopedilum*, subsidiary cells were recognized by Rosso (1966) but not by Faber (1904) or by Rutter and Willmer (1979). This need not reflect differences in the material studied. Rather, it reflects the subjective nature of subsidiary cell definitions and the differing criteria used for their recognition.

The criteria most commonly used are those of cell configuration and cell sizes. Zörnig (1904) described a species of *Dendrochilum* in which the whole stomatal complex, including four subsidiary cells, is no larger than an average epidermal cell. Although subsidiary cells are often markedly smaller than averge epidermal cells, this is an extreme case.

Lateral and polar subsidiary cells often differ with respect to size, and the lateral ones tend to be more easily recognized. When they arise as perigene cells (see discussion on development), the lateral cells often manifest themselves by the disorder they create in the epidermal cell files. This criterion seems to be the one most extensively used by Williams (1976, 1979). Perhaps this is why the stomatal complexes depicted in these reports are in many cases interpreted as having two lateral subsidiary cells only, whereas according to the illustrations they could just as well be described as consisting of two polar cells in addition to the two lateral ones.

This is mentioned merely to demonstrate how difficult the judgment of subsidiary cells can be. Descriptive terms such as "paracytic" and "tetracytic" may nevertheless be useful when it is recognized that these terms inevitably force a continuum of forms into discrete categories.

Other criteria by which subsidiary cells can be recognized are surface features and contents. Zörnig (1904) observed that the subsidiary cells of some species of *Coelogyne* project more from the surface than other epidermal cells. Cuticular patterns may also help to distinguish cells in close association with guard cells as subsidiary cells. Both Zörnig (1904) and Sprenger (1904) observed cuticular rims enclosing the whole stomatal complex.

Very prominent subsidiary cells were noted by Pridgeon and Williams (1979) and Pridgeon (1982) in *Dresslerella* and some species of *Pleurothallis*. In these plants, the subsidiary cells elevate the stomatal apparatus above the epidermal surface.

Subsidiary cells were distinguished by the absence of (calcium oxalate?) crystals in an otherwise crystal-containing epidermis of *Vanilla* (Roux, 1954; Nayar et al., 1976). Zörnig (1904) noted oil droplets both in subsidiary cells and in guard cells in one species of *Coelogyne*, whereas oil seems to have been absent in epidermal cells.

Subsidiary cells have been observed in all three families of Orchidales. In the Cypripediaceae, the reports are somewhat contradictory. Faber (1904) did not mention any, and Rutter and Willmer (1979) did not see them, but they were observed by Rosso (1966) in two genera. In Orchidaceae, subsidiary cells are now known in the Neottioideae, Epidendroideae, and Vandoideae, but apparently they never occur in the Orchidoideae. It seems that the presence of subsidiary cells is very common in Orchidales (see Table 1 in Williams, 1979) and that this condition is more widespread than the absence of subsidiary cells, that is, anomocytic stomata, although this condition is also common.

Structure in Cross Section

Orchid stomata generally are flush with the epidermis (Möbius, 1887; Tominsky, 1905; Cyge, 1930; Solereder and Meyer, 1930). Some species have slightly raised stomata: Bletiinae, especially *Calanthe* (Fig. 4-2-I; Möbius, 1887); some species of *Coelogyne* (Zörnig, 1904); *Eria muscicola* (Lindl.) Lindl. (Fig. 4-2-H) and *Malaxis latifolia* J. E. Sm. (*Microstylis congesta*, Tominsky, 1905); and a few species of Pleurothallidinae (Pridgeon and Williams, 1979). On the basis of studies of tropical and temperate orchid species, respectively, Tominsky (1905) and Ziegenspeck (1938) suggested that this condition may be related to a humid habitat.

Slightly sunken stomata in some species of *Paphiopedilum* and *Phragmipedium* (Faber, 1904; Rosso, 1966) result mainly from a lowering of the surrounding epidermis (Ziegenspeck, 1938). This is probably also the case with the species of *Palumbina, Odontoglossum* (Ayensu and Williams, 1972), *Trichopilia,* and *Helcia* (Möbius, 1887). Only a single instance of very sunken stomata has been described in orchids, namely in *Ponera striata* Lindl. (Fig. 4-2-F; Möbius, 1887).

The rarity of sunken stomata in orchids is surprising (Tominsky, 1905), considering the frequent occurrence of other xeromorphic stomatal characters. An explanation, however, may be sought in purely morphogenetic factors during stomatal development, as suggested by Ziegenspeck (1938); see later discussion of development.

Stomatal Ledges

Among the xeromorphic traits often encountered in orchid stomata are the well-developed outer cuticular ledges (Fig. 4-2; see, however, the later discussion of function). Inner cuticular ledges are often absent or only weakly developed (Solereder and Meyer, 1930). The outer ledges are often curved and almost completely enclose a pitcher-shaped outer chamber. In some cases, the inner walls of the outer chamber bulge out from the two sides thus narrowing the middle of the cavity into a slit between an upper and a lower space (Fig. 4-2-G).

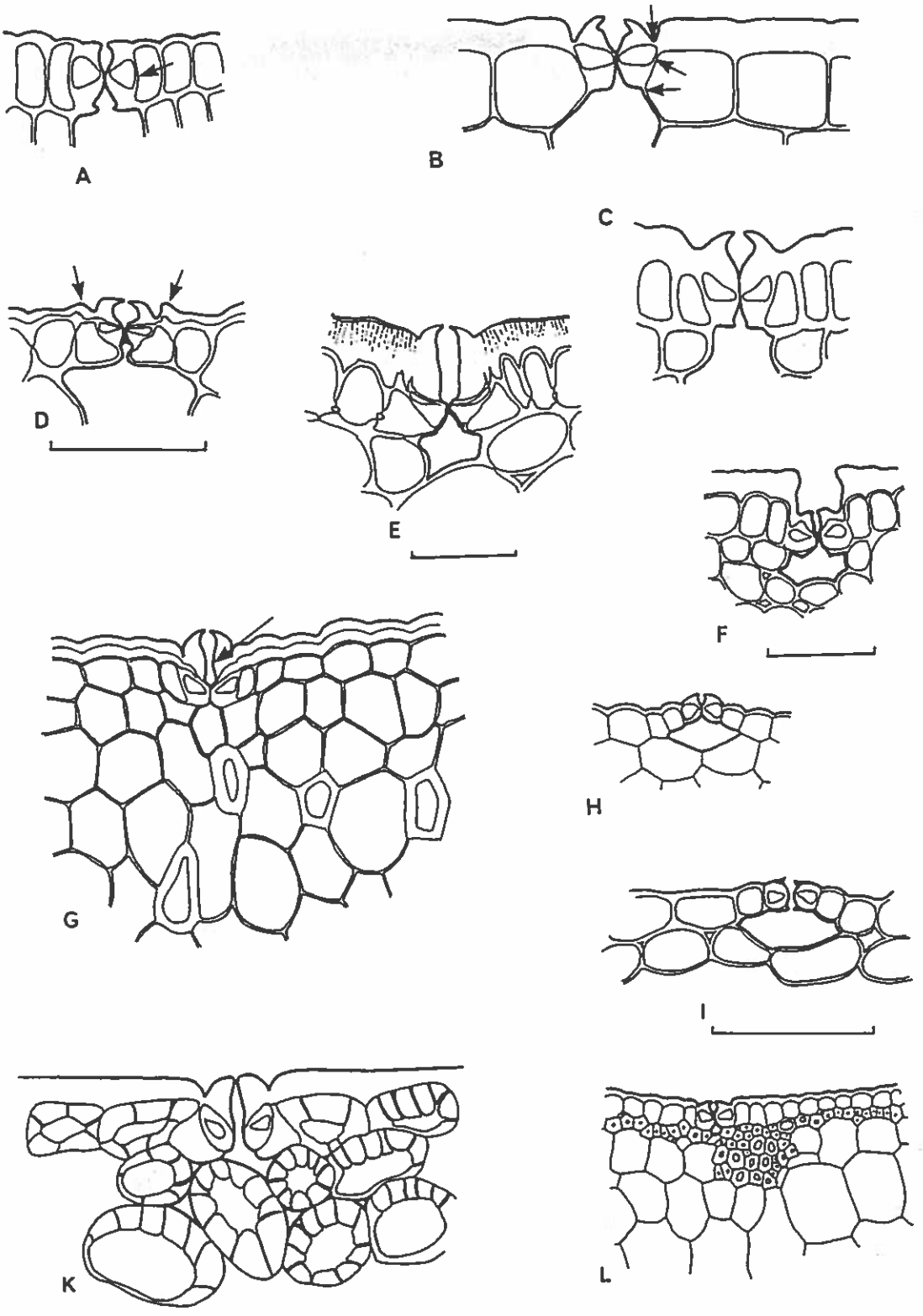
The outer ledges unite at the polar end of the stoma to form a ring, collar, or an almost complete roof above the stomata, as described earlier.

Cell Wall Thickening

The guard cell walls are often heavily thickened, especially in species from dry habitats (Tominsky, 1905; Solereder and Meyer, 1930).

The pattern of unequal thickening of guard cell walls is thought to have important implications for guard cell movement, as discussed by Schellenberg (1896), who included two kinds exemplified by orchid species. The true identity of the first example, called *Cymbidium aloifolium,* is doubtful, however (Seidenfaden, 1983). *Cymbidium aloifolium* (Fig. 4-2-A) belongs to the *Amaryllis*-type (Schwendener, 1881), which is characterized in cross section by a triangular cell lumen with almost equal sides, one corner facing toward the stomatal pore between the outer and the inner ledge. *Paphiopedilum insigne* (Lindl.) Pfitz. (Fig. 4-2-B; *Cypripedium insigne,* Schellenberg, 1896) belongs to the *Helleborus*-type, in which the guard cell lumen is an obtuse-angled triangle. *Maxillaria picta* W. Hook. (Fig. 4-2-C) also conforms to the latter type, although Schellenberg (1896) considered the stomata nonfunctional because of their heavily thickened walls. Mohl (1856) depicted stomata that resemble the *Amaryllis*-type in *Dactylorhiza majalis* (Rchb.) Hunt and Summerh. (*Orchis latifolia*), but they have an almost circular lumen in cross section. The guard cell walls are almost evenly thickened, except for the ledges and a thinner area toward the flanking epidermal cells. A similar structure was described by Siebe (1903) in Apostasiaceae.

The condition found in *Eria muscicola* (Fig. 4-2-H; Tominsky, 1905) differs slightly from the *Amaryllis*-type. Here, one corner of the almost isosceles triangular lumen faces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the lower part of the stomatal pore and there is no inner ledge. This resembles the structure of stomata in certain, mainly hygrophytic, dicotyledons (Guttenberg, 1959). Hence, yet another stomatal type can be assigned to the orchids.

Substomatal Chamber

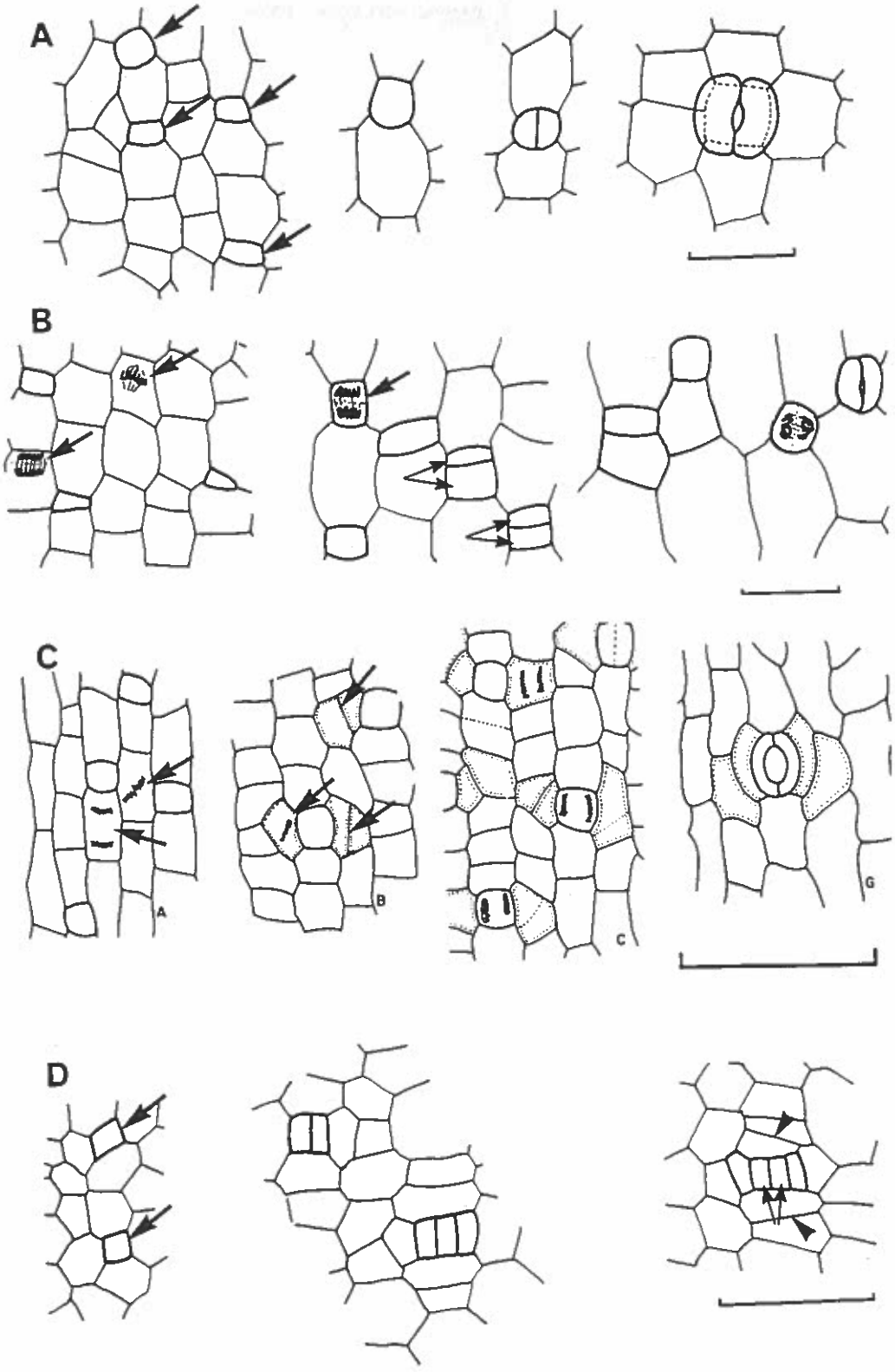
The substomatal chamber in orchid stomata often appears to be very narrow (Fig. 4-2). Furthermore, Sprenger (1904), Zörnig (1904), and Tominsky (1905) noted that the tiny substomatal chambers of some species were lined with schlerenchymatous mesophyll (Fig. 4-2-K, L). Zörnig noted U-shaped wall thickenings of the cells with the thickened part adjacent to the substomatal chamber in *Coelogyne punctulata* Lindl. (*C. ocellata* Lindl.). The air stream from such stomata into the mesophyll is confined to narrow intercellular passages (Zörnig, 1904). This is seemingly a very inefficient circulation system, but it probably reduces stomatal transpiration. It would be very interesting to know the exact conditions under which such stomata occur and function. A correlation between small chamber size and xeric habitat was found by Tominsky (1905).

Development

Possibly the very first account of stomatal development was given by Mohl (1838) in *Hyacinthus orientalis* L. The sequence of events, as observed by Mohl, starts with the stomatal meristemoids, as they are now called, which divide parallel to the longitudinal axis of the leaf to form the guard cells. These guard cells later separate along their common wall to form the stomatal pore. The origin of the meristemoids, however, was not described in this early work.

The first complete account of this agenous stomatal development was given by Strasburger (1866–67), who observed the unequal mitoses in protodermal cells which lead to the differentiation of meristemoids. Among the many species with this kind of development, *Dactylorhiza majalis* (Rchb.) Hunt and Summerh. (*Orchis latifolia*) was the only orchid mentioned (Fig. 4-3-A). Strasburger mentioned casually that *Orchis latifolia* resembled other local German species ("überhaupt unsere einheimische Orchideen"), indicating that he had studied quite a few. Later studies in Orchidoideae concern *Orchis militaris* L. (Ziegenspeck (1944b), *Habenaria marginata* Coleb. (Inamdar, 1968), and several other genera in Orchidinae (Rasmussen, 1981a). These contributions establish the agenous stomatal development as the predominant one in this group.

Figure 4-2. Orchid stomata in cross section. **A.** *Cymbidium aloifolium* "*Amaryllis*-type" of stoma. **B.** *Paphiopedilum insigne* (Wall.) Pfitz. (*Cypripedium*) "*Helleborus*-type" of stoma. "Hinges" indicated with arrows, see text for description. **C.** A presumably functionless stoma with heavily thickened guard cell walls, *Maxillaria picta* Hook. **D.** Cuticular rims (arrows) above the subsidiary cells, *Bulbophyllum coriaceum* Ridl. ex Stapf. **E.** Extreme development of cuticula and a very narrow guard cell lumen, *Sarcanthus rostratus* (Lodd.) Lindl. **F.** Stomata sunken in relation to epidermal cells, *Ponera striata* Lindl. **G.** Well-developed cuticular ledges that almost divide the outer cavity into two parts, *Luisia zeylanica* Lindl. **H.** *Eria muscicola* (Lindl.) Lindl. and **I.** *Calanthe masuca* (D. Don.) Lindl., stomata slightly raised above the epidermis. **K.** *Bulbophyllum vaginatum* (Lindl.) Rchb. f. (*Cirrhopetalum whiteanum* Rolfe) and **L.** *Cymbidium bicolor* Lindl., substomatal chamber very narrow and surrounded by sclerenchymatous tissue. Scales: D and I = 0.10 mm; E–F = 0.05 mm. No scales indicated in originals of other figures. (Sources: A–C, Schellenberg, 1896; D–F and I, Möbius, 1887; G, H, and L, Tominsky, 1905; K, Sprenger, 1904.)


A
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Mesogene Cells

Another kind of stomatal development that occurs concurrently with the agenous developmental pattern is also observed in Orchidoideae. It is the hemimesogenous pattern (Rasmussen, 1981a), which involves an additional developmental step after meristemoid formation (Fig. 4-3-B). The meristemoid divides once, producing a mesogene cell and a meristemoid of second order, which eventually divides into guard cells.

This hemimesogenous kind of stomatal development was also observed in Neottioideae, namely, *Ludisia discolor* (Ker-Gawl.) A. Rich. and *Spiranthes* sp. (Williams, 1975). This was the first study in which mesogene cells in monocotyledonous plants were recognized.

Both Williams (1975) and Rasmussen (1981a) noted isolated occurrences of mitoses in cells adjacent to the guard cell mother cell. Such divisions by definition lead to perigene cells, but consistent patterns of perigene cells such as those found in Epidendroideae and Vandoideae (Williams, 1979) have not been observed in Neottioideae or in Orchidoideae.

A common feature of the hemimesogenous developmental pattern in orchidoid as well as neottioid orchids seems to be the consistent position of the single mesogene cell on the side of the guard cells toward the leaf base. In contrast, Singh (1981) observed mesogene cells formed at the longitudinal side of the meristemoid in several species of *Coelogyne* and *Dendrobium*. In some instances, Singh described two mesogene cells flanking the guard-cell mother cell. As many as three mesogene cells, completely surrounding the guard-cell mother cell were observed in *Pholidota articulata* Lindl. var. *griffithii* K. and P. (eumesogenous development; Singh, 1981).

The most complex developmental pattern as yet reported for orchid stomata is that of *Vanilla planifolia* Jacks. ex Andr. and *V. wightiana* Lindl. ex Hook. f. (*V. wightii* Lindl.; Nayar et al., 1976). In these species the meristemoid divides parallel to the leaf axis to produce a lateral mesogene cell as well as a meristemoid of second order (Fig. 4-3-D). This meristemoid subsequently divides, forming a meristemoid of third order, which is surrounded by two opposing mesogene cells. The last meristemoid eventually forms two

Figure 4-3. Surface views of stomatal development in orchids. **A.** Agene development. Left: stomatal meristemoids (arrows) appear as minor cells in the protodermis. Middle: the meristemoids grow into an isodiametric shape and divide longitudinally. Right: eventually, the two guard cells part forming the stomatal pore. *Dactylorhiza majalis* (Rchb.) Hunt and Summerh. (*Orchis latifolia*). **B.** Agene and hemimesogene development. Left: stomatal meristemoids arise by unequal divisions (arrows) of protodermal cells. Middle: some meristemoids divide transversely (arrow) to form a set of an apical meristemoid and a basal mesogene cell (double arrows). Right: meristemoids divide longitudinally to form guard cells. *Orchis canariensis* Lindl. **C.** Hemiperigenous development. Left: divisions around meristemoids produce perigene cells (arrows), which may subdivide (middle, arrows), mainly the lateral ones. The meristemoids divide longitudinally into guard cells and eventually separate to form the stomatal pore (right). Perigene cells of mature complex indicated by stippling. *Oncidium flexuosum* Sims. **D.** Eumesoperigenous development. Left: meristemoids are seen as minor, isodiametric cells (arrows). Middle: meristemoids divide longitudinally several times. Right: meristemoid has divided three times; the two central cells (arrows) constitute the guard cells, flanked by two lateral, presumably mesogene cells. Divisions in adjacent epidermal cells result in polar perigene cells (arrowheads). *Vanilla planifolia* Andr. Scale: 0.05 mm. All details are oriented with tip of leaf toward top of page. Note that mitoses support the interpretation of the development only in B and C, and that the starting point for the description is earlier in B than in A, C, and D. (Sources: A. Strassburger, 1866–67; B, Rasmussen, 1981a, courtesy of *Botanical Journal of the Linnean Society;* C, Williams, 1979, courtesy of *Botanical Journal of the Linnean Society;* D, Nayar et al., 1976. A redrawn from original, B drawn from photos, D redrawn and reoriented according to the authors' descriptions.)

guard cells. Before that, however, the epidermal cells abutting the meristemoid divide into perigene cells. The guard cells are flanked by two mesogene cells and at the polar ends by perigene cells, hence the development can be expressed as eumesoperigenous. In *V. wightiana* the surrounding cells are often subdivided further, either tangentially or radially. Variation in this developmental pathway does exist, and in the development of some stomata only one mesogene cell is formed; in other cases there may be three. This developmental pattern, which is outstanding in orchids and in monocotyledons generally, deserves additional study, especially for assessment of the actual mitoses taking place; the article by Nayar et al. (1976) shows no mitotic figures. N. H. Williams (personal communication) has observed a perigenous stomatal development in species of *Vanilla*.

Mesogene cells in many cases defy observation at maturity, and hence we cannot possibly tell how frequently they occur in the numerous orchid species that have not been investigated developmentally. Mature epidermal patterns offer guidance, though. Patterns that resemble those of *Ludisia discolor*—for instance, *Zeuxine strateumatica* (L.) Schltr. according to Williams (1975), *Beadlea elata* (Sw.) Small (*Spiranthes elata* [Sw.] L. C. Rich.) according to Dressler (1981, fig. 3.11), and the patterns recorded as anisocytic by Lavarack (1971)—might involve mesogene cells, but developmental studies are needed to assess this.

Perigene Cells

Various perigenous patterns are common in Orchidales, although this has only been appreciated recently (Williams, 1976, 1979). The patterns are diverse, but their common feature is that the epidermal cells adjacent to the meristemoid divide to produce a special configuration of cell walls in the vicinity of the future guard cells (Fig. 4-3-C). Most often, walls formed in the laterally abutting cells are oblique to the longitudinal axis of the future pore.

Strasburger (1866–67, p. 326) saw the perigenous development in some unspecified "tropische Orchideen." Which tropical orchids Strasburger studied cannot be ascertained, except for the fact that in his summary (p. 335) *Stanhopea tigrina* Batem. is mentioned, and in a description of wall thickenings of guard cells he referred to *"Stellis pulchella."* (Presumably, *"Stellis"* is a misspelling of *"Stelis."*) In his description, it is mainly the laterally adjacent cells that become divided, often in a distinctly oblique fashion, forming trapezoid or triangular lateral perigene cells. The development is similar to that described for *Aloe* (illustrated in table XL, 111–112 of Strasburger, 1866–67). Strasburger's mention of this developmental pattern in orchids is so brief that it escaped the attention of later reviewers (Bary, 1877; Paliwal, 1969). Williams (1976) studied the development of stomata in 15 species of Cymbidieae (including Gongoreae) and Maxillarieae. Usually two oblique mitoses occur in each laterally neighboring cell, the two new cell walls being nonintersecting. A trapezoid cell results, which may again divide parallel to the guard-cell mother cell. Occasionally, polar perigene cells are also observed (Fig. 4-3-C). In this feature the species differ, but the difference is insufficient to distinguish between the tribes and subtribes studied. An estimation of the frequency with which perigene cells are formed and subdivided will have to rest largely on each worker's

personal judgment of cell configuration. Another vandoid tribe, Oncidieae (here included in Cymbidieae as Oncidiinae), was found to agree well with the ones mentioned earlier (Williams, 1979). This general developmental pattern is also found in *Bulbophyllum* (*Cirrhopetalum*, Epidendreae) according to Singh (1981).

Occurrence in Orchidales

The stomatal development in Orchidales shows no less diversity than that found generally among monocotyledons. The eumesoperigenous kind described by Nayar et al. (1976) in *Vanilla* and the eumesogenous kind reported by Singh (1981) in *Pholidota* both are unique in monocotyledons and deserve further confirmation. The hemimesogenous development first observed by Williams (1975) was later found in other orchids (Rasmussen, 1981a) as well as in other members of Liliiflorae (Conover, 1982; Rasmussen, 1983). The hemiperigenous or euperigenous development with oblique divisions occurring lateral to the meristemoid has been described several times in orchids (Strasburger, 1866–67; Williams, 1976, 1979) and similar developmental patterns are well known from many monocotyledons (Tomlinson, 1974).

Conover's study (1982) indicated a close association of unusual kinds of stomatal development with net-veined monocotyledon leaves. It is tempting to connect the diversity in the stomatal development of orchids with their large variation in leaf shapes. Still, most orchids have not been studied and additional patterns may emerge. A survey of the Orchidales (Williams, 1979) clearly showed the paucity of available information on stomatal development in certain parts of Orchidales.

Differentiation and Morphogenesis

In angiosperms, stomatal development is generally assumed to begin with an unequal division of an epidermal cell, by which the stomatal meristemoid is produced (Tomlinson, 1974; Rasmussen, 1981b). Yet this kind of origin of the meristemoids is not mentioned in several references (Inamdar, 1968; Nayar et al., 1976; Williams, 1976, 1979), perhaps because of the use of inappropriate material for these studies (as discussed by Rasmussen, 1981b). Nevertheless, the matter deserves critical attention because lack of unequal division in some stomatal developmental pathways would challenge our conception of the differentiating effect of this kind of mitosis.

It has been stressed on several occasions that the developmental events are not always apparent from the mature apparatus (Paliwal, 1969; Tomlinson, 1974). The morphogenetic processes that shape the maturing guard cells and their immediate surroundings are of decisive importance for mature structure and function. Most ontogenetic studies mentioned earlier focus on the sequence of mitoses leading to the formation of the guard cells, and as such the studies are essentially two-dimensional in their approach. Ziegenspeck's work (1938), however, is concerned with the events at right angles to the surface. In meristemoids and in cells adjacent to them, the cell wall reinforcement is oriented in such a way as to restrict their elongation. This arrangement has a major impact on the three-dimensional structure of the maturing apparatus. The stomatal meristemoid in some monocotyledons has the shape of a frustum of a four-

sided pyramid, with sloping anticlinal walls. In *Allium*,[1] for instance, the flat top of the pyramid points out of the leaf, and hence the meristemoid tends to sink between the epidermal cells when they enlarge (Ziegenspeck, 1938). In orchids, the meristemoid is shaped similarly, but the truncated top of the pyramid points into the leaf, so the meristemoid tends to protrude above the surface when the epidermal cells stretch. The orchid species studied by Ziegenspeck (1938) were all native to Germany. The same sloping of the dorsal walls can be seen in numerous illustrations of mature stomata in orchids from other parts of the world, however, which indicates that it may be a widespread orchid feature (Fig. 4-2). Ziegenspeck's interpretation is in harmony with the fact that hardly any sunken stomata are found in this plant group, although other xeromorphic stomatal characters prevail.

Another important finding of Ziegenspeck concerns the changing birefringence patterns during stomatal ontogeny. The first published observations of this nature (Ziegenspeck, 1936) deal with European terrestrial orchids, but the principles later proved applicable to several other monocotyledons (Ziegenspeck, 1944b). In the young protodermal surface of an orchid leaf, the orientation of microfibrils in the cell walls is generally at right angles to the long axis, as judged by studies with a polarization microscope. This texture facilitates a longitudinal stretching of protodermal cells, and cell divisions occur mainly in transverse direction (Ziegenspeck, 1936, p. 599). In the meristemoids, the microfibril orientation at first resembles that of other protodermal cells, but later the orientation becomes longitudinal, and the same applies to the proximal part of the laterally abutting protodermal cells. After the longitudinal division of the meristemoid into guard cells, another change of predominant microfibril orientation takes place, this time producing a pattern radiating from the future pore (Fig. 4-4). In poorly developed stomata, this radiating pattern may exist without the formation of a pore, but a pore is never developed if the radiating microfibril orientation fails to be established (Fig. 4-4-E–G). This suggests a causal connection between changes in the cell wall texture and the formation of the pore.

The most striking features of the mature stomatal apparatus are the cuticular ledges and surface patterns around the guard cells. These specializations are probably often responsible for the recognition of the developmentally associated (mesogene and perigene) cells as subsidiary cells. Where such features are lacking, the associated cells tend to resemble other epidermal cells. The processes that take place during the differentiation of the subsidiary cells and their various distinctions remain to be studied. Also the final stretching of the epidermal cells has an important impact on the appearance of the mature stomatal complexes (Rasmussen, 1983).

Distribution

Stomata may be found on all plant organs except for roots (Willmer, 1983). This fact is strikingly demonstrated in orchids. It should be remembered that in epiphytic orchids

[1]Not an orchid.—Ed.

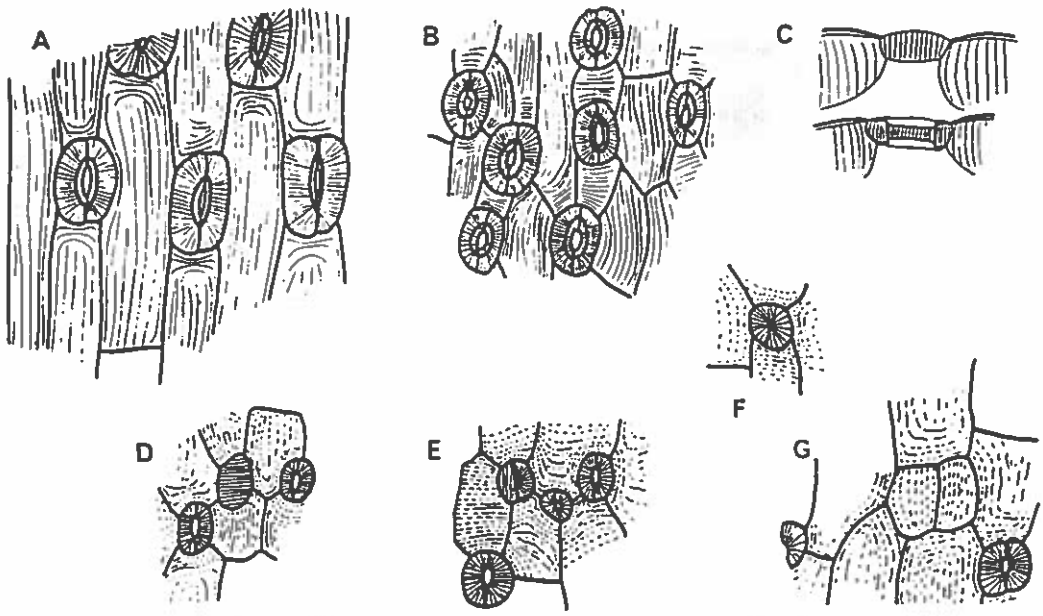

Figure 4-4. Microfibril orientation in cell walls of stomatal apparatus and surrounding epidermal cells, according to studies with polarization microscopy. A–B. Normal stomata in surface view, microfibrils of the periclinal guard cells walls radiating from the pore. In adjacent epidermal cells they are generally at right angles to those of the guard cells. *Anacamptis pyramidalis* (L.) Rich. (A) and *Himantoglossum* cf. *hircinum* (L.) Spreng. (B). C. Stoma in longitudinal section, tangential (above) and equatorial (below), showing anticlinal orientation of microfibrils in anticlinal walls. *Orchis militaris* L. D–G. Stomata abnormally developed or arrested in development. One meristemoid, probably arrested shortly after the unequal mitosis, has transverse microfibril orientation (D). Another has reached the stage of longitudinal microfibril orientation and has divided longitudinally (E, upper left), but only one of the resulting guard cells has acquired the radiating microfibril orientation, and a pore has not formed. *Ophrys fuciflora* (F. W. Schmidt) Moench. F–G. Other irregularities are the attaining of radiating microfibril orientation before the meristemoid has divided into guard cells (F) and arrested development of guard cells before radiating microfibril orientation was attained (G). In both cases no pore is formed. *Orchis militaris* L. (Source: A–G, Ziegenspeck, courtesy of Eugen Ulmer publishers; no magnifications indicated in original.)

all organs, including the roots, are exposed to light. Some orchids lack real leaves and, apart from an occasional inflorescence with a number of bracts, they consist entirely of the photosynthesizing roots. Even in such orchids, exemplified by species of *Campylocentrum* and *Taeniophyllum* (Porsch, 1908), no stomata occur on the roots. Another kind of opening, or pneumathode, functions as an aeration system on roots with photosynthetic activity (Schimper, 1884).

Stomata occur infrequently on rhizomes and stems (Weltz, 1897, a survey of 130 species; Solereder and Meyer, 1930). Inflorescence axes have stomata more often than do stems (Hering, 1900, a comparison of 50 orchid species). Stomata on inflorescence axes and on floral bracts were thought by Ziegenspeck (1936) to stimulate transpiration and thereby to increase nutrient supply to the flowers and fruits. On the other hand, Sprenger (1904) did not find stomata on inflorescences, pseudobulbs, or rhizomes in Bulbophyllinae. Faber (1904) observed stomata on stems in species of *Paphiopedilum* and *Phragmipedium*. These stem stomata had comparatively less developed ledges and a larger cell lumen than those found in the leaves. This finding suggests that the stem

stomata were nonfunctional. Rosso (1966) saw stomata in the leafy stems in *Cypripedium* and *Selenipedium* but considered them to be rare on inflorescence axes. In species of *Vanilla,* stomata were found on stems, leaves, and fruits (Roux, 1954), the latter also observed by Gertz (1919). Hew et al. (1980) found stomata in several floral parts of orchids, such as sepals, petals, even labellum and column. F. Rasmussen (personal communication) observed stomata on the inner epidermis of several orchid fruits.

Some stomata observed in organs other than foliage leaves seem to be rigid and thus functionless, as is strongly indicated, for example, by Hew et al. (1980). Others would be expected to function normally in larger, green organs such as the *Vanilla* fruits. It is surprising, however, that the green, long-lived pseudobulbs of Bulbophyllinae seem to photosynthesize without stomata on their surfaces (Sprenger, 1904). An interesting exception is presented by *Bulbophyllum minutissimum* (F. Muell.) F. Muell. and another, similar species (Pfitzer, 1884) in which a cavity develops in each of the tiny pseudobulbs. The bottoms of such cavities are lined with stomata, which open into a dense mesophyll.

Hypostomaty—Amphistomaty

The stomata of orchid foliage leaves are mostly confined to the lower surface. The surveys by Möbius (1887), Lavarack (1971), Williams (1979), and Avadhani et al. (1982) all show a distinct predominance of hypostomatic over amphistomatic leaves. This has been confirmed in studies of more restricted orchid groups: Cypripediaceae (Faber, 1904), Orchideae (Rasmussen, 1981a), Bulbophyllinae (Sprenger, 1904), and Pleurothallidinae (Pridgeon, 1982). Epistomatic leaves have not been observed (Chatin, 1856, confirmed by all subsequent examinations).

The amphistomatic condition may be more or less pronounced. In some cases it amounts only to a few scattered stomata on the upper leaf surface, as in *Calanthe brevicornu* Lindl. (Singh and Singh, 1974). Amphistomatic species seem to fall into two categories. One category comprises terrestrial, hygrophytic species, such as *Dactylorhiza incarnata* (L.) Soo (*Orchis incarnatus,* Solereder and Meyer, 1930). The other comprises succulent-leaved epiphytes. Of course, hypostomaty is also sometimes found in species of these categories. Möbius (1877) found stomata on both sides on the thick leaves of Oncidiinae: *Oncidium* spp. and *Trichocentrum albo-coccinium* Lindl. (*T. albopurpureum* Rchb. f.); Laeliinae: *Cattleya* spp.; and Vandeae: *Chamaeangis odoratissimum* (Rchb. f.) Schltr. (*Angraecum odorantissimum* Lindl.), *Aerides multiflorum* Roxb. (*A. affine* Wall.), *Adenoncos virens* Bl. (*Aerides virens* Lindl.), and *Rhynchostylis retusa* Bl. In contrast, the amphistomatic leaves in Cypripediaceae were found in the most hygrophytic of the three genera studied (Faber, 1904), that is, *Cypripedium.* These observations agree with the general pattern in other angiosperms, namely, that hypostomaty is most frequent in mesophytic species, whereas amphistomaty is most frequent in very dry and humid habitats (Parkhurst, 1978). In terete or ensiform leaves, stomata naturally occur on the whole surface. An additional factor related to stomatal distribution is the existence of Crassulacean acid metabolism (CAM, discussed later) in many orchids. This pathway of carbon fixation is correlated with thick leaves (Mott et al., 1982), which are associated with amphistomaty.

Tominsky (1905) studied orchid species from two different tropical vegetation types: an arid shrub forest where epiphytes are rare and a tall, dense, humid forest. Amphistomaty was mostly found in species from the arid forest area. Tominsky considered this a surprising result, because it seemed unlikely that species with many xeromorphic characteristics would have an increased number of stomata. In fact, this need not be the case. As shown by Rousteau (1981) for Cactaceae, amphistomaty can be associated with a smaller total number of stomata than hypostomaty. Tominsky suggested a causal relation between a more or less vertical orientation of the leaves and amphistomaty, however. Cyge (1930), who noted amphistomaty in mesophytic and hygrophytic European terrestrial orchids with upwardly pointed leaves, shared Tominsky's opinion.

Density

De Bary (1877) assessed the normal range of stomatal density in higher plants to be from about 40 to 300 stomata per square millimeter. The figures calculated for orchids are within the lower part of this range (17–121/mm^2, Ziegenspeck, 1936; 40–110/mm^2, Singh and Singh, 1974; 8–180/mm^2, Avadhani et al., 1982). The lowest frequency was noted in the most succulent leaves in the species studied by Goh et al. (1977). Ziegenspeck (1936) found higher densities primarily in species from marshy habitats.

Nayar et al. (1976) noted an increasing stomatal index (number of stomata in relation to number of epidermal cells) from the base toward the apex of the leaf in *Vanilla*. Variations in stomatal density within the plant can also be observed in European terrestrial orchids (Cyge, 1930). In these latter species, the number of stomata per unit surface area is always higher in upper leaves than in lower leaves on the same stem. This finding was interpreted as due to the stronger light intensity on the upper leaves. The notion that light intensity could be positively correlated with density of stomata is supported by comparisons between orchid species with different preferences for shade (Cyge, 1930). One could object to comparisons of a single feature between distantly related species (such as Orchidoideae with Neottioideae). Cyge's suggestion, however, is supported by experiments with dicotyledonous species, in which the stomatal index increases in response to stronger light intensity (Schoch et al., 1980).

Function

Mechanics of Stomatal Movement

The opening of stomata as a result of increased turgor depends very much upon the radial microfibril orientation of the guard cell walls (Aylor et al., 1973). One of the first accounts of this general feature of guard cells is based on orchid material (Ziegenspeck, 1936). This extensive work is mainly known for the vast amount of information it conveys on the life histories of European terrestrial orchids. It also contains a section devoted to stomata of the "Serapideen," an aggregate of genera containing *Orchis* (including *Dactylorhiza*) and *Ophrys* (Ziegenspeck, 1936, pp. 594–606). Similar but less de-

tailed accounts of stomatal mechanics in orchids were published later (Ziegenspeck, 1938, 1944b).

Stretching of the cell wall occurs most readily at right angles to the microfibrils. In mature guard cells, as seen in a surface view, microfibrils are predominantly oriented radially in relation to the open pore (Fig. 4-4-A, B; Ziegenspeck, 1936). and in the anticlinal walls they are oriented at right angles to the leaf surface (Fig. 4-4-C). Hence, the microfibril pattern is generally perpendicular to the long, curved axis of the guard cells. The flanking epidermal cells are characterized by a longitudinal microfibril orientation, perpendicular to that of the guard cells. In response to increasing turgor, the guard cells yield mostly in the longitudinal direction. This elongation is mainly on the outer part of the guard cells, as the areas around the pore are reinforced by the stomatal ledges. The antagonistic microfibril orientation of the flanking epidermal cells tends to impede an increase in the total length of the guard cells, whereas it easily gives way to the sideward pressure caused by an increased curvature of the guard cells that will open the pore.

This sideward bulging of guard cells during opening differs somewhat according to the structure of the guard cells. Generally, the guard cell lumen tends toward circularity in cross section when the turgor rises, but in most guard cells the structure is modified by differential wall thickenings. Schwendener (1881) defined different kinds of stomata on the basis of the positions of narrow cell wall parts as seen in transection, which are the "hinges" (Hautgelenke) about which guard cells move. This classification of stomata has been applied to certain orchid species (Schellenberg, 1896). In *Cymbidium aloifolium* (*Amaryllis*-type, Fig. 4-2-A) the adjacent epidermal cells have thin walls toward the guard cells and toward the leaf surface and therefore readily yield to a movement of the guard cells parallel to the epidermal surface.

The lumen of guard cells in *Paphiopedilum insigne* (*Helleborus*-type, Fig. 4-2-B) show an obtuse-angled triangle in cross section, not an isosceles triangle as in *Cymbidium*. With rising turgor, the lumen approaches an isosceles triangle in shape, that is, the obtuse angle is reduced. When the outer walls of the subsidiary cells are thick, only the lower part of the guard cells can move. In such cases the hinges are very distinct. The outer cavity is little changed by the movement, but the inner cavity enlarges and the guard cells bulge against the flanking subsidiary cells. The mobility of guard cells is impeded by strong wall thickenings, such as in old leaves of *Maxillaria picta* (Fig. 4-2-C). A variation of the *Helleborus* type (Guttenberg, 1959) is similar but with the ventral corner of the triangular guard cell lumen pointing toward the inner cuticular ledge instead of facing the middle of the ventral wall. This condition is indicated in *Eria muscicola* according to the illustration by Tominsky (1905); see Figure 4-2-H. The opening movement of such a stoma is supposed to occur more obliquely than in the usual *Helleborus*-type (Guttenberg, 1959).

Actual measurements of normal stomatal movements were performed by Mohl (1856) on *Gymnadenia conopsea* (L.) R. Br. and by Ziegenspeck (1936) on *Ophrys fuciflora* (F. W. Schmidt) Moench and *Orchis militaris* L. Both workers found conditions resembling the one that Schellenberg (1896) called *Amaryllis*-type, but the lumen of the guard cells is nearly circular in cross section. Mohl (1856) found that the widening of the pore was

mainly caused by a change in shape of the lumen of the guard cells in cross section from circular to elliptical, so that the dorsal walls did not move perceptively. The stomatal movements measured by Ziegenspeck (1936) included the dorsal walls and were greater at the outer surface of the guard cells than within. Experiments with stomatal movements under various conditions were performed by Müller (1872) in *Herminium monorchis* (L.) R. Br.

Subsidiary Cells

The function of the cells adjacent to the guard cells depends on their antagonistic mechanical properties, as mentioned earlier. Turgor pressure of the guard cells must increase beyond a certain point before the adjacent cells give way for the opening of the pore. On the other hand, the resistance due to wall structure also contributes toward closing the stoma, when turgor in the guard cells decreases (Ziegenspeck, 1936).

In osmotic experiments with many species, Mohl (1856) observed very different stomatal responses depending on whether the subsidiary cells were intact. In the orchid species studied (among others, *Gymnadenia conopsea, Listera ovata* [L.] R. Br., *Cypripedium calceolus* L.) the movements of the stomata were only slightly affected when the subsidiary cells had been damaged.

The fact that subsidiary cells are often partly suspended over the substomatal chamber is assumed to enhance their flexibility, so that they may act as an "elastic buffer" (Meidner and Mansfield, 1968). Because of their position, subsidiary cells also constitute the link between guard cells and the rest of the plant in regard to transport of water and substances (Maier-Maercker, 1979). In orchids, however, there are examples of substomatal chambers so narrow that the subsidiary cells cannot be said to be suspended (Fig. 4-2), although they do connect the guard cells with other tissues.

It may be noted that the functional peculiarities suggested here apply to all cells adjacent to guard cells with no reference to their ontogenetic origin. The association of the developmentally related cells (that is, the perigene and the mesogene cells) with the function of the mature stomatal apparatus remains uncertain.

Transpiration

Many of the features characterizing orchid stomata may be said to be decidedly xeromorphic. The outer cuticular ledges are often well developed, especially in succulent leaves (Krüger, 1883), guard cell walls are often thickened, and substomatal chambers are narrow and occasionally lined with sclerenchyma. Haberlandt (1892) noted the seeming paradox that many plant species of the tropical rain forest exhibited such characteristics, even though the transpiration was low compared to temperate conditions. An explanation was sought in the very high light intensities, which for a short period of the day are assumed to critically raise the plants' demand for water. Such "temporary drought" conditions might be especially relevant for epiphytes.

With this in mind, it is surprising that the epiphytic orchid included in Haberlandt's study, *Grammatophyllum speciosum* Bl., did not show distinct xeromorphic traits except for

some succulence in the stem. Haberlandt found later (1904) in the tropical-forest terrestrial *Paphiopedilum venustum* (Wall.) Pfitz. (*Cypripedium*) large stomatal ledges, which supposedly reduce stomatal transpiration by enclosing a small chamber of humid air. On the other hand, Tominsky (1905) reported xeromorphic characteristics such as large cuticular ledges and narrow substomatal chambers in species naturally occurring in a seasonally dry habitat. Epiphytic orchid species from a humid forest habitat did not show these characteristics, although well-developed stomatal ledges occur in temperate terrestrial orchids (Ziegenspeck, 1936), even in species confined to marshy habitats. Here the function of the large stomatal ledges was interpreted not as part of xeromorphy but as a protection of the pore against blocking by dew. Functional explanations may be offered for large stomatal ledges in all habitats.

On the whole, however, the xeromorphic condition of many epiphytic orchid stomata seems well established on the basis of characteristics such as the thickness of cell walls and cuticula, as well as the features of the substomatal chamber. The frequent succulence of the plants and the general absence of stomata on stems and pseudobulbs probably also reflect a necessity to economize on water. The same hypothesis applies to the occurrence of CAM in orchids (Nuernbergk, 1963).

Rhythms

Many orchids, mainly epiphytes, have been shown to assimilate CO_2 via CAM (the most recent list is found in Avadhani et al., 1982). In such plants the rhythm of stomatal movement is reversed. C_3 and C_4 plants open their stomata during the day and close them during the night; CAM plants open their stomata at night, when the temperatures and the transpiration rates are low. The CO_2 is fixed and malic acid accumulates during the night. The following day while the stomata are closed, CO_2 is released and refixed via the C_3 pathway.

Physiological studies of the opening and closing of stomata in orchids by Borriss (1967) and Goh et al. (1977) showed that individual species differ considerably in their opening and closing times. Moreover, species that have not been reported to have CAM (such as certain temperate terrestrial orchids, *Gymnadenia conopsea* [L.] R. Br. and *Dactylorhiza sambucina* [L.] Soó [*Orchis sambucina*]) may be found to have open stomata in the evening and during the night (Leitgeb, 1886; Stahl, 1919). Such species probably acquire a minor part of their CO_2 by dark fixation, as is the case in *Cattleya* (Knauft and Arditti, 1969).

A structural requirement for CAM seems to be succulence (Nuernbergk, 1961). In a number of orchids, Neales and Hew (1975) showed a correlation between leaf thickness and CAM. The CAM-succulence is often but not always associated with amphistomaty (Borris, 1967), and succulent orchids with CAM are not structurally very different from succulent species without CAM (Nuernbergk, 1961). There seem to be no detectable structural differences between CAM-stomata and others. The specific responses of stomata in connection with CAM have attracted comparatively little attention in the rather comprehensive literature on the CAM pathway itself.

Energy Supply

The movement of stomata is considered to be an energy-dependent process. Three sources of energy for guard cells have been suggested: oxidative phosphorylation, photophosphorylation, and a blue-light photoreceptor system (Zeiger, 1983). The importance of these sources in individual species may differ, as indicated, for instance, by studies of *Paphiopedilum* stomata. It has been shown that species of *Paphiopedilum* lack chlorophyll in their guard cells (Nelson and Mayo, 1975; Rutter and Willmer, 1979). This is an exceptional case (the first and only, according to Rutter and Willmer, 1979), in which photophosphorylation can be disregarded as an energy source. The *Paphiopedilum* stomata responded normally to several agents affecting stomatal movement, such as blue light and CO_2 levels (Nelson and Mayo, 1975). It was also shown that plants of *Paphiopedilum insigne* have very low rates of stomatal conductance and of photosynthesis (Williams et al., 1983). New findings about stomatal function in *Paphiopedilum* have important implications for stomatal physiology in general.

The absence of starch in stomata of *Allium cepa* L. renders it an interesting species for anatomical and physiological comparisons with species that have the usual starch content in guard cells. For this reason, *Allium* stomata have been studied closely. Examination revealed an alternative reserve carbohydrate system based on soluble fructans (Allaway, 1981). Several orchid species are also reported to lack starch in their stomata, as mentioned earlier, and the same or other alternative carbohydrates may be involved here. Therefore, morphological and physiological studies of these and other orchids would be quite useful.

Stomata and Saprophytism

Studies of vegetative structure in the holosaprophytic orchid species began very early (Lory, 1847) and these species have attracted considerable attention. Most orchid species have been described as more or less dependent on a mycorrhizal relationship. The holosaprophytic species represent an extreme case of fungus dependency, whereas green orchids require mycorrhiza only during the earliest stages of life.

The function of stomata in CO_2 uptake for photosynthesis is obviously negligible in nongreen plants, but their role in the regulation of transpiration may be just as important as in green ones.

Stomata in green orchids are distributed mostly on the lower surfaces of leaves and rarely on stems and inflorescences. This is not the case in the saprophytic species. Reduction in aerial organs would seem to be a logical characteristic for the holosaprophytic life form and has been observed often. The leaves (which are often rudimentary) carry few stomata, but stomata are regularly found on stems, inflorescence axes, and even rhizomes. It is typical that in a study of stem anatomy (Möbius, 1886) the only mention of stomata occurs in connection with the saprophyte *Limodorum abortivum* (L.) Sw., which has many stomata on the stem. In rhizomes of *Corallorhiza odontorhiza*

Nutt., elevated stomalike structures, interpreted as water pores, with large "substomatal chambers" were observed by Holm (1913). Similar structures were noted in *Epipogium aphyllum* Sw. (Porsch, 1905) and in *Hexalectris spicata* (Walt) Bark. (Fig. 4-5-C; *Corallorhiza arizonica* Wats., MacDougal, 1899). Stomata are found on scape and scale leaves of *Aphyllorchis pallida* Bl. (Groom, 1895). In *Lecanorchis malaccensis* Ridl. they are found mainly on subterranean scale leaves, and decrease in number in the upper part of the scape (Groom, 1895).

The appearance of stomata in these positions varies, but they rarely resemble the functional stomata of green species. Those observed by Groom (1895, p. 18) in *Aphyllorchis* and *Lecanorchis* were "raised on slight eminences and the guard cells protrude" (Fig. 4-5-F). The guard cells may be unequal, or the apparatus may appear to be torn out of position obliquely or transversely (Fig. 4-5-A, F; Porsch, 1905, *Neottia nidus-avis* (L.) L. C. Rich.; Groom, 1895, *Aphyllorchis*). The closing of such stomata is mechanically impossible. Another frequent abnormality is the incomplete development of ledges, such as in the stem of *Limodorum abortivum* Sw. (Fig. 4-5-D; Möbius, 1886), which could also affect the ability of guard cell movement. Finally, there are stomata with pores that seem never to have opened or to have become partly or completely blocked (Fig. 4-5-E; Lory, 1847; Porsch, 1905). On *Neottia* stems, Porsch (1905) noted stomata with normally developed ledges and substomatal chambers, but the ventral walls of their guard cells were connected (Fig. 4-5-B).

It is interesting to note that the development of stomata resembling those on saprophytic orchids could be induced experimentally by subjecting parts of autotrophic plant species to high humidity and darkness (Wassermann, 1924). Saprophytic species often grow in shady conditions. Therefore, it is possible that the abnormal appearance of their stomata could be partly due to environmental factors, especially for stomata in subterranean positions on a rhizome.

In a number of saprophytic orchids, the search for stomata has proved negative (*Pogoniopsis* sp., *Wullschlaegelia aphylla* Rchb. f. [Porsch, 1905], *Galeola javanica* (Bl.) Benth. and Hook., and *Epipogium roseum* [D. Don] Lindl. [Groom, 1895, the latter as *E. nutans* Rchb. f.]).

Two evolutionary trends apparently govern the stomatal apparatus of saprophytic orchids. The first involves the formation of stomata that are unable to close, possibly functioning as water pores; the second is the development of stomata that are permanently closed, which have no function. As a final step in the evolution of stomata in saprophytic orchids we may see the complete loss of that structure.

Taxonomy and Phylogenetic Considerations

No paper on stomatal morphology has been cited and criticized more frequently than that by Stebbins and Khush (1961). These authors attempted to bring the morphology of stomata in monocotyledonous plants into a phylogenetic and taxonomic context. The aim was laudable, but the foundation was very narrow, as noted by later critics (Tomlinson, 1969, 1970, 1974; Williams, 1975). Stebbins and Khush selected four unspecified

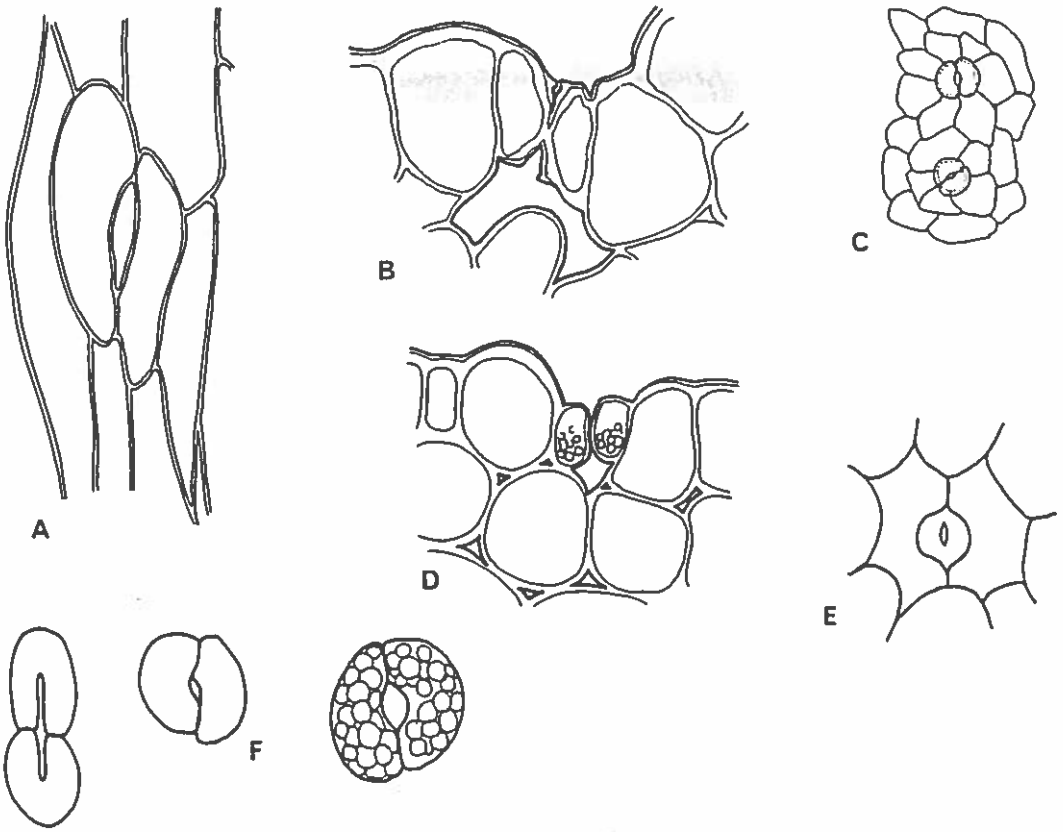

Figure 4-5. Stomata in species of saprophytic orchids. **A.** Surface view. **B.** Cross section of stomata with guard cells distorted or not properly separated. *Neottia nidus-avis* (L.) Rich., from scape. **C.** Stomata from "coralloid branches" on rhizome of *Hexalectris spicata* (Walt.) Bark. **D.** Cross section of stoma with a very small substomatal chamber and lacking guard cell wall thickenings, stem of *Limodorum abortivum* (L.) Sw. **E.** Surface view of structure resembling a stoma, but with a cutinized obstacle in the pore so that this is reduced to a narrow slit. Interpreted as a functionless, reduced stoma. Rhizome of *Epipogium aphyllum* Sw. **F.** Surface view of one distorted and two seemingly normal stomata from scape and scale leaves in *Aphyllorchis pallida* Bl. (Sources: A–B, Porsch, 1905; C, MacDougal, 1899; D. Möbius, 1886; E–F, Groom, 1895. No scales indicated in originals. All redrawn.)

orchids, plus one member of Apostasiaceae (not included in the orchids) to represent all orchids (a plant group that comprises more than a third of all monocotyledonous species). Earlier literature was referred to only to a very limited extent, and the conclusions (at least those concerning orchids) were at variance with well-established facts.

As a result, the study of Stebbins and Khush (1961) presents a very incomplete picture regarding orchids. This fact was not obvious to all orchidologists, however: "Stebbins and Khush (1961) have convincingly shown that the stomatal complex in the Apostasioideae is anomocytic, i.e., two guard cells without subsidiary cells which is characteristic of the Orchidales" (Garay, 1972, p. 202). Withner, Nelson, and Wejksnora (1974) also found the evidence of Stebbins and Khush convincing, although the former authors referred several times in their 1974 paper to Solereder and Meyer (1930), whose work could have led to a revision of their view if they had studied the illustrations. Figure 28

(Solereder and Meyer, 1930) is a line drawing of the stomatal complex in *Liparis viridiflora* (Bl.) Lindl. (*L. longipes*), showing four distinct subsidiary cells.

Stebbins and Khush's conclusion entered higher-order systematics, probably through the forementioned sources, and appeared in such works as Cronquist (1968, p. 322, discussion of Liliidae), who stated that "the stomates are without subsidiary cells or (in a few small families) have two subsidiary cells," and Huber (1977, p. 291), who wrote, "the Liliiflorae are almost sufficiently described by possession of anomocytic stomata." These examples show how suppositions tend to gain ground by uncritical repetition without further studies. Several papers now effectively refute the statement that orchids lack subsidiary cells (Williams, 1975, 1976, 1979). Referring to Solereder and Meyer (1930), Williams (1976) stated that the subsidiary cells in orchids were known but neglected for more than forty years. Actually, the first information about them can be traced back to Möbius (1887). It is now generally accepted that within the orchids there are many kinds of stomata, both with respect to number of subsidiary cells and with respect to their developmental origin.

This notion is reflected in the attempts to use stomatal characters at lower levels in orchid systematics. Many papers on orchid systematic anatomy include keys, but stomatal features generally play a minor role. Möbius (1887) used the amphistomatic condition of some species as a diagnostic character. Rosso (1966) utilized the same character on the generic level in Cypripediaceae. Zörnig (1904) distinguished *Pleione* from the other Coelogyninae by its lack of subsidiary cells. Stomatal size, which is sometimes very small in Coelogyninae, could be used to distinguish species within *Dendrochilum* (*Platyclinis*) and *Coelogyne*. In similar studies, stomatal characters were of little help to the systematics (Faber, 1904; Hünecke, 1904; Sprenger, 1904). Aoyama and Tanaka (1980), however, claimed that they could identify an intergeneric hybrid between a *Cymbidium* and a *Bifrenaria* through stomatal morphology.

In phylogenetic discussions it is most important to evaluate whether similar character states are phylogenetically homologous, and to formulate a feasible hypothesis of the history (that is, the phylogenetic polarity) of each character.

Often a more detailed study will expose character states that resemble each other superficially but are not directly connected historically. Ontogenetic studies have been used to break up assumed homologies in stomatal characters. For instance, it can be shown by ontogenetic studies that the paracytic condition comprises at least three independent character states in monocotyledons (Tomlinson, 1969) when ontogeny is considered. The origin of the paracytic condition, therefore, cannot be treated as a single evolutionary event in the monocotyledons.

Even when there is no reason to doubt the unique origin of character states, their evolutionary polarity is often difficult to assess. However, one example of a stomatal character that may be useful in phylogenetic reconstruction is presented by *Dresslerella* and a few species of *Pleurothallis* (Pridgeon, 1982). In these species the subsidiary cells elevate the whole apparatus above the epidermal level in a very characteristic fashion. Several methods to polarize character states exist, but only criteria based on out-group comparison are logically sound (Stevens, 1980; Watrous and Wheeler, 1981). The elevated stomata of *Dresslerella* may exemplify in a simple way this procedure with the

phylogenetically well-defined group of Pleurothallidinae as the taxon being examined and all other orchids constituting the out-group. Because elevated stomata occur only within Pleurothallindinae, we may assume that this character state is derived from stomata that are flush with the epidermis, a condition found both in Pleurothallidinae and in the out-group. The genus *Dresslerella* and a small part of *Pleurothallis*, characterized by the derived state, constitute a likely monophyletic group in Pleurothallidinae.

In order to polarize the character known as "number of subsidiary cells" within the Orchidales, Williams (1979, p. 64) used a criterion of character correlation: "Within the epidendroid orchids generally, the more primitive members lack subsidiary cells and the more advanced members possess subsidiary cells." On this basis, the condition of several subsidiary cells was presupposed to be the derived character state in Epidendroideae (Williams, 1979). The argument rests on the assumption that primitive character states in different characters occur in correlation with each other. The degree to which derived or primitive character states are correlated, however, depends on the evolutionary history of the group in question (Stevens, 1980). Advanced stomata could, for instance, easily occur in taxa primitive in other respects. Indeed, the widespread distribution in both diandrous and monandrous orchids of subsidiary cells suggests that the presence of many subsidiary cells is ancestral in orchids generally. This would seem to weaken Williams's conclusions concerning their evolutionary polarity in Epidendroideae.

Studies of plant structures often include phylogenetic considerations that are unclear because the above-mentioned problems are neglected. It is hoped that present and future attention to phylogenetic homology and polarization of character states will provide us with a far better foundation for understanding the diversity in stomatal structure as well as the evolution of other characters.

Glossary

Agene cell. A cell that abuts the stomatal meristemoid and was present before its formation.
Agenous stomatal development. The future guard cells are completely surrounded by agene cells.
Amphistomaty. The condition of having stomata on both sides of the leaf.
Anisocytic stomatal complex. Mature guard cells surrounded by subsidiary cells of unequal size.
Anomocytic stomatal complex. Mature guard cells surrounded by epidermal cells not deviating morphologically from other epidermal cells.
Anticlinal. At right angles to the surface of an organ; often used to describe the orientation of a new wall (opposed to periclinal).
Apical. Facing toward the apex of the leaf (for instance, apical perigene cells as opposed to basal; Fig. 4-6).
Basal. Facing toward the leaf basis (as opposed to apical; Fig. 4-6).
CAM. A set of chemical reactions through which atmospheric CO_2 is fixed into a C_4-compound (malic acid) which accumulates during the night and is later transferred to the Calvin cycle during the day while the stomata are closed.
Cyclocytic stomatal complex. Mature guard cells surrounded by an undetermined, often larger number of similar subsidiary cells radiating from the circumference of the guard cell pair.
Diacytic stomatal complex. Mature guard cells sorrounded by a pair of subsidiary cells with their common walls at right angles to the long axis of the guard cells.
Ensiform. Shaped like a two-edged sword.
Epidermis. Superficial cell layer of plants in which the stomata are formed.

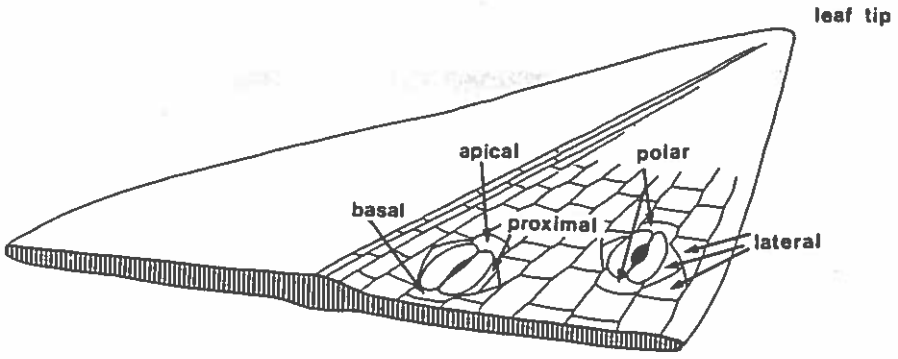

Figure 4-6. Vignette.

Eumesogenous stomatal development. The stomatal meristemoid becomes completely surrounded by mesogene cells.
Eumesoperigenous stomatal development. The stomatal meristemoid becomes completely surrounded by mesogene and perigene cells.
Euperigenous stomatal development. The stomatal meristemoid becomes completely surrounded by perigene cells.
Floating stomata. Mature guard cells surrounded by a single, ring-shaped cell (a condition encountered in some species of ferns).
Frustum. The part remaining when the top is removed from a pyramid or a cone with a horizontal cut.
Guard-cell mother cell. A cell that will go through a symmetrical mitosis producing a guard cell pair.
Hemimesogenous stomatal development. The future guard cells partly surrounded by mesogene cells, the rest of the surrounding cells being agene.
Hemimesoperigenous stomatal development. The future guard cells surrounded by mesogene, perigene, and agene cells.
Hemiperigenous stomatal development. The future guard cells partly surrounded by perigene cells, the rest of the surrounding cells being agene.
Holosaprophyte. A plant that is unable to photosynthesize and is totally dependent on decaying organic material for its survival.
Hygromorphic feature. Feature typical of a hygrophyte.
Hygrophyte. A plant growing in humid (hygric) conditions (not to be confused with hydrophyte, an aquatic plant).
Hypostomaty. The condition of having stomata only on the lower side of the leaf (as opposed to epistomaty).
Lateral. Toward the edges of the leaf (for instance, lateral perigene cells, opposed to polar; see Fig. 4-6).
Longitudinal. Parallel to the long axis of the leaf (in a longitudinal division the resulting wall is longitudinal).
Mesogene cell. A cell arising from a division of a stomatal meristemoid and which is not itself a stomatal meristemoid or a guard cell.
Mesophyll. Photosynthesizing ground tissue of a leaf located beneath the epidermis.
Mesophyte. A plant growing in intermediate (mesic), that is, not in any way extreme, climatic conditions.
Microfibril. Strand of cellulose in the plant cell wall.
Ontogeny. The growth and development of an organism to maturity, or any part of this process.
Paracytic stomatal complex. The mature guard cells surrounded by two flanking subsidiary cells.
Periclinal. Parallel to the surface of an organ. Often used to describe the orientation of a new cell wall (as opposed to anticlinal).
Perigene cells. Cells arising during stomatal development by the division of protodermal cells around the stomatal meristemoid.
Phylogeny. The process and history of evolutionary branching.

Polar. Toward the basal and apical ends of the leaf (for instance, polar perigene cells as opposed to lateral ones; Fig. 4-6).
Protodermis. Epidermis in a very early stage, in which cells proliferate by mitoses.
Proximal. Toward the center of the stomatal complex (for instance, proximal perigene cells, as opposed to the farther ones; Fig. 4-6).
Sclerenchyma. Tissue with heavily thickened cell walls.
Stoma (pl., stomata). An elliptic pore in the epidermis and two specialized kidney-shaped epidermal cells, the guard cells, which surround it. By their movements the two guard cells can regulate the size of the pore, and hence the ventilation of the leaf mesophyll.
Stomatal meristemoid. A cell formed by an unequal division of a protodermal cell, or of another stomatal meristemoid. A cell that can give rise to a pair of guard cells.
Subsidiary cell. Epidermal cell adjacent to the mature guard cells and structurally deviating from other epidermal cells.
Terete. Circular in cross section, tapering or narrowly cylindrical.
Tetracytic stomatal complex. Mature guard cells surrounded by two polar and two flanking subsidiary cells.
Transverse. At right angles to the long axis of the leaf (in a transverse division the resulting wall is transverse).
Xeromorphic feature. Feature typical of a xerophyte.
Xerophyte. A plant growing in very dry (xeric) conditions.

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