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Autor(en): **Huynh, Kim-Lang / Sampson, F.B.**

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Flower structure in *Freycinetia banksii* (Pandanaceae) of New Zealand

Kim-Lang Huynh [1] **and F. B. Sampson** [2]

[1] Phanerogamy Division, Botanical Institute, University of Neuchâtel, CH-2007 Neuchâtel, Switzerland
[2] School of Biological Sciences, Victoria University of Wellington, Wellington, New Zealand

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Abstract

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In *Freycinetia banksii* each pistillate flower bears staminodes and each staminate flower has a pistillode. Both vary widely in composition (2–18 carpels, 1–16 staminodes and 4–13 carpellodes, 5–13 stamens in the flowers studied). They have basically the same numbers of male components (stamens/staminodes) and female components (carpels/carpellodes) with the former components generally in larger numbers. The most frequent numbers of male components are 7–11 and female components 6–9. Some unusual anatomical features are observed. The pistillode has some lateral vascular bundles corresponding to those in the pistil (in other species, lateral vascular bundles were absent in the pistillodes). A 2-layered parenchyma tissue, bordered at either side by 2–4 layers of sclerenchyma, is observed along the central region of the intercarpellary partitions in the upper part of the pistil (such parenchyma was not found in other species). Further methods for studying the flower structure in *Freycinetia* are described.

Keywords: anatomy, carpellode, flower structure, *Freycinetia*, *F. banksii*, methodology, New Zealand, Pandanaceae, pistillode, staminode, taxonomy.

Introduction

Floral structure in *Freycinetia banksii* Cunn., the only New Zealand species, was studied to enable comparison with other species of the genus. Warburg (1900: 40–41) observed 8–12 stamens and "several stigmas with variable numbers". Moore & Edgar (1970: 98) reported that the staminate spikes were "quite hidden by tightly packed fls. ♂ of several stamens ... surrounding a small rudimentary ovary; limits of individual fls not easily determined. ♀ with 6–12 purplish staminodes ...; stigmas c. 6–12 ...". The essence of this description was repeated in Moore & Irwin (1978): "A male floret consists of a few stamens and a rudimentary ovary, ...; each flattened ovary has a ring of staminodes ...". Stone (1973: 242) also found that "the number of stigmas ... varies roughly between 6 and 12".

These numbers of 8–12 stamens and 6–12 staminodes, however, need confirmation. Given the fact that flowers of both sexes are strongly congested, it is most probable that such numbers have been obtained by separating flowers along pistils/pistillodes and counting the male components (stamens/staminodes) still adherent to these (see also Warburg 1900: Fig. 6G and Moore & Irwin 1978: Fig. 1c). This was used in the past for obtaining numbers of male components in flowers. It may provide some rough idea of floral structure, but can be misleading. In fact, in transverse sections of some separated pistils of *F. banksii* some adherent staminodes were not associated with the pistil observed, their xylem being clearly centrifugal to it. In other words, this xylem was centripetal to the nearby removed pistil and, consequently, those staminodes were in reality associated with the latter pistil (see below). In addition, in species where flowers are strongly congested, two nearby male components each belonging to a different flower may coalesce along their filaments, as observed in *F. reineckei* Warb. by Huynh & Cox [Flower structure and potential bisexuality in *Freycinetia reineckei* (Pandanaceae), a species of the Samoa Islands – in press in Bot. J. Linn. Soc.]. Consequently, the male components of such a pair are inseparable and may remain adherent to one and the same female organ when the flowers are separated.

It follows that the flower of *F. banksii* should be studied anatomically, using the method described in Huynh (1991: 313–315). This method has proved suitable for any species of *Freycinetia*. With this species, an anatomical study of the staminate flower can also establish the structure of the pistillode, in particular the carpellode number.

There are other reasons too for this anatomical study. As seen in Warburg (1900: 40) and Moore & Edgar (1970: 98), the pistil of *F. banksii* has a widely variable carpel-number which may be the highest in *Freycinetia* (this may be the case in *F. baueriana* Endl., of Norfolk Island, as well). Its staminate flower, too, probably has a widely variable stamen-number which may be the highest in the genus since a correlation seems to exist between the carpel and the stamen numbers in *Freycinetia* (Huynh 1991: 326). Consequently, *F. banksii* is probably the most interesting example for studying this variation. As to the highest number of stamens in *Freycinetia*, its knowledge is of evident interest in any study of evolution in the family Pandanaceae: for example, for comparison with *Pandanus*, in which the stamen number may be up to "many hundreds", as observed in *P. lamprocephalus* Merr. & Perry (St. John 1973: 56).

The primary aim of the present paper is to study the numerical structure of the flower of *F. banksii*, i.e. its carpel/carpellode and staminode/stamen numbers. Some other anatomical features seem useful to enable further comparisons with other species or in future taxonomic revisions of *Freycinetia*, and are reported too.

With regard to its taxonomic status, *F. banksii* shares some features with *F. baueriana*. Both have more or less the same stigma numbers. Stone (1973: 242) considered them conspecific and assigned subspecific rank to the former, viz. *F. baueriana* Endl. ssp. *banksii* (Cunn.) Stone. Their differences seemed weak, being essentially based on the colour of the flowering bracts (deep salmon-pink in *F. baueriana*, white or white tinged with purple in *F. banksii*). Consequently, the subspecific status for *F. banksii* "may be open to question" (Stone 1973: 243–244). Given this uncertainty, the specific rank of *F. banksii* is maintained in the present paper, while keeping in mind the "need to seek some additional evidence and to evaluate bract color in terms of its significance" (Stone, pers. comm. 9 March 1992). Both species, indeed, need further study. Thus, the pistillate spikes of *F. baueriana* were described as solitary by Warburg (1900: 29), but in the Kew Herbarium an infructescence collected by Pat Ralston in July 1965 showed 4 spikes. As to *F. banksii*, both its pistillate and staminate spikes were solitary according to Warburg

(1900: 29); however, the pistillate inflorescences studied in the present paper comprised 3 or 4 spikes and the staminate inflorescences 4–6 spikes. Also, Sampson when collecting material for the study found staminate inflorescences to commonly have 5 or 6 spikes. He further corroborated Moore & Edgar's observation (1970: 98) that plants of either sex had a similar stem diameter (about 4 cm near their base). On the contrary, pistillate shoots of *F. cumingiana* Gaudich. cultivated in Hamburg were about 15–18 mm in diameter but staminate ones were only about 5–6 mm (Poppendieck 1987: 314 and 315, and Fig. 3e).

Material and techniques

Pistillate spikes at anthesis and early fruiting stages and staminate spikes at anthesis were used. All were fixed either in FAA or in 70% ethanol as soon as collected by Sampson either in December 1990 or February 1991, in a native forest at Belmont Reserve, eastern Hutt Hills, Lower Hutt, North Island, New Zealand, approximately 41°11′44″S 174°52′25″E.

For light microscopy, part of the material was microtome-sectioned and stained with combined aqueous safranin and astra blue. Fruiting spikes were used for studying the numerical structure of the pistillate flower. In fact, when observed from above, pistils were clearly distinct on these (see also Moore & Irwin 1978: Fig. 1d). Therefore, it was possible to count stigmas and select flowers with various stigma numbers for that study. In contrast, pistils were indistinct on spikes at anthesis; and these were used to study the pistil structure at that stage. With fruiting spikes, however, thorough dehydration of pistils prior to embedding in paraffin was rarely possible in entire pistils, which generally caused sections of the lower part to be broken. Since that part, and not the upper part, was essential for study of the numerical structure of the flowers, the upper parts of the pistils were removed before dehydration in absolute ethanol. As for staminate flowers, they were also indistinct. Given the wide variation in size of these flowers, and the fact that their longer axes are more or less parallel to the spike axis (Fig. 19) – as are those of the pistillate flowers (Moore & Irwin 1978: Fig. 1d) –, fractions of about 18 × 5 mm cut along the middle part of spikes were used. That part is the only one that may provide large fractions with a more or less plane surface, which is the primary condition for optimal tangential sections. Since such sections contained a large number of flowers, it was necessary to be able to recognize any flower for further checking of data observed. For this purpose, the sections were divided into three equal parts by small marks using red coloured cosmetic nail polish at both margins on cover slips. The positions of the flowers studied were found by referring to these parts and also to their carpellode numbers (for example: slide 12, section 6, middle part, upper-left flower with 9 carpellodes). Furthermore, in order to avoid accidental use of flowers that might have been partly amputated at one end when portions of spikes were removed for sectioning, all the peripheral flowers, either partial or apparently entire, in transverse sections were disregarded.

Another part of the material was used for scanning electron microscopy (SEM) with a Philips PSEM 500 after being critical-point dried with CO_2 and sputter-coated with ca. 400 Å gold. Sections in some preparations examined by light microscopy were also investigated by SEM. For this purpose, the preparations were immersed overnight in toluol to remove the cover slips and clean the sections from the synthetic mounting resin. They were then passed through absolute ethanol, after which squares of 14 × 14 mm or less containing the sectioned material were excised with a glass-cutter, passed through acetone and dried by the critical-point method. They were then mounted on stubs and subsequently examined in the scanning electron microscope after coating. The sections of pistillate flowers were previously drawn with a camera lucida for further identification on SEM micrographs of the staminodes they bore (e.g. Fig. 7). Pollen grains too were investigated by SEM.

Phloroglucinol-HCl stain was used to identify lignin in stamens, staminodes, carpels and carpellodes (tracheary elements; endothecial thickenings; other lignified elements). The identification was made either by staining free-hand sections or by squashing material in the stain on a slide.

A method for identifying the male components in the flower of *Freycinetia banksii*

It was more difficult to determine the number of male components per flower in *F. banksii* than in other species of *Freycinetia*, for two reasons. Firstly, although this flower was more or less discernible in its basal outline, just beneath the pistil/pistillode cavity – especially in pistillate flowers (Fig. 7 and 8: the upper and lower flowers) –, that base usually occupied two or three consecutive sections, which did not facilitate the study. In fact, its anatomical elements at the same level were generally not all in the same plane, given both its large width, currently up to 9 mm or more, and the curvature of the spikes. Secondly, the number of male components varied widely and sometimes unpredictably (see Tables 1 and 2). This required great caution when identifying male components and rendered the study more difficult by requiring repeated and time-consuming verifications. Without previous experience obtained by the first author when studying other species where flowers were less congested and male components were fewer (e.g. *F. cumingiana*, *F. javanica* Blume var. *expansa* Stone, *F. reineckei*, *F. scandens* Gaudich., etc.), the present study would have been very difficult to undertake.

In *F. cumingiana* (Huynh 1991), for example, the simplest case in *Freycinetia*, the flowers have 2 female components (carpels/carpellodes) and 2 or 3 male components (staminodes/stamens). Transverse sections of their bases showed that the flowers were sufficiently distinct (for pistillate flowers) or very clearly distinct (for staminate flowers) from one another (Huynh 1991: Fig. 4 and 14). The basal region of such a flower was formed of a female organ and of 2 or 3 vascular bundles around it, each of these corresponding to a male component. The xylem in these vascular bundles was centripetal, i.e. more or less oriented towards the central part of the flower. Particularly the staminate flowers were very distinct when observed from above, each having 2 or 3 stamens all clearly located within a distinct floral enclosure.

The fact that each male component has only one vascular bundle and that the xylem in this vascular bundle is centripetal to the flower, has been observed in all the species studied. These two features are the main criteria for identifying male components in any *Freycinetia* flower (Huynh 1991: 313). The "centripetalism" concept should be emphasized. In *F. cumingiana* where the basal transverse section of the flower is more or less isodiametric, the xylem in the male components is oriented towards the centre of the flower or a point near it (Huynh 1991: Fig. 1 and 4). But with the strongly elongated flower of *F. banksii* it is also the strongly elongated central part of the flower – not the centre as a point – that the "centripetalism criterion" should be applied to. That is to say: only the xylem in the male components on or near the apices of the axes of the flower is oriented towards the centre of this, while the xylem in the others is opposite to the longer axis (Fig. 4). Furthermore, at upper levels where they are free, stamen/staminode filaments may be subjected to some twisting by nearby filaments or pistillodes/pistils, or may become curved. It follows that whenever used, the "centripetalism criterion" should in the first place be applied to the flower bases which are always located beneath the spike surface.

The "one whorl criterion" was found in the course of the present study and also used: the flower of *Freycinetia* has only one whorl of male components (see also Huynh 1991: 325–326). It may be applied in other species with strongly congested flowers. In *F. banksii* it was frequently applied. An example may be shown by the three staminodes more or less in the same row between the three pistils around the centre in Fig. 7. The left staminode may be assumed to be associated with the left pistil

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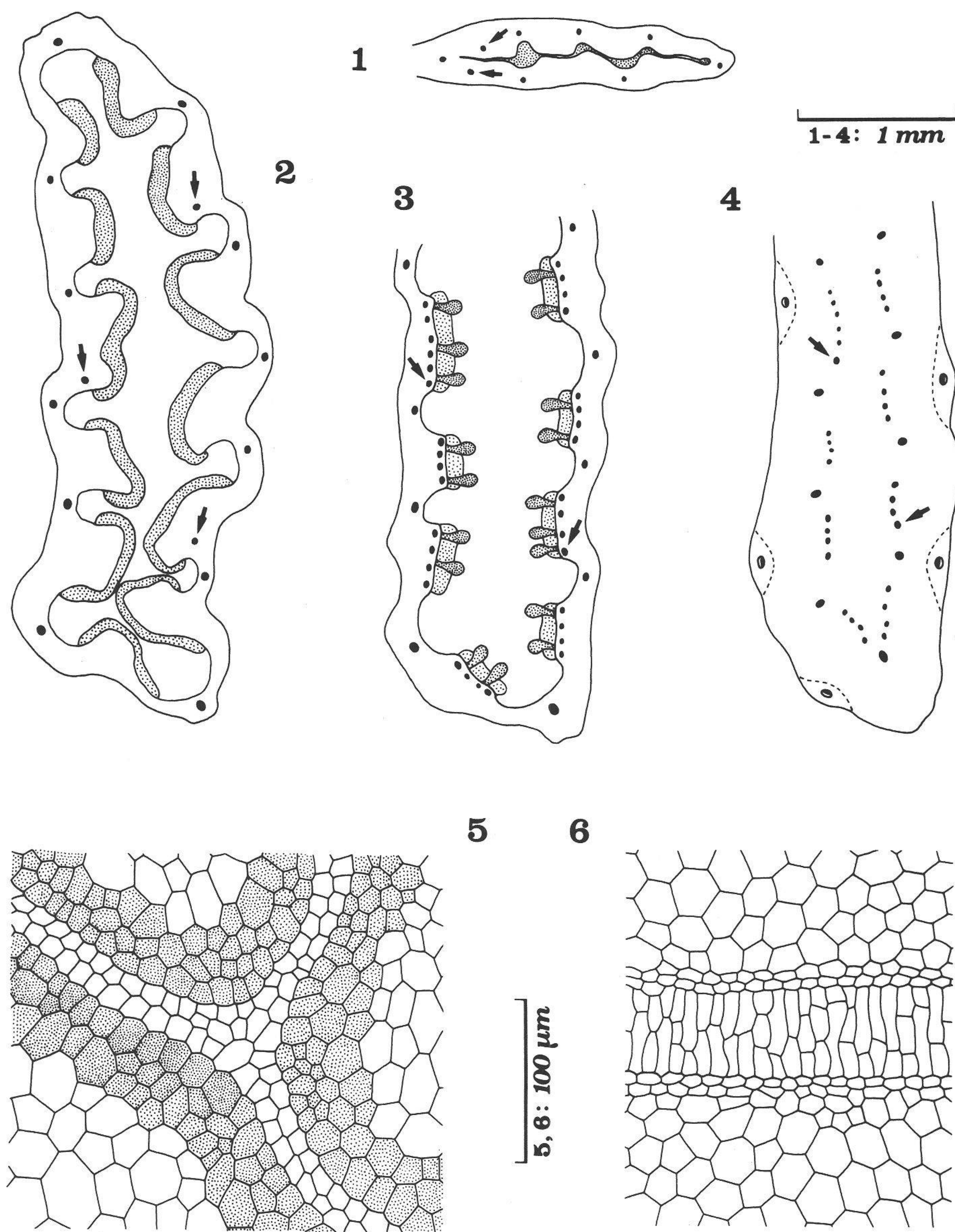
Fig. 1–6. *Freycinetia banksii*. – Fig. 1: transverse section of pistillode with 7 carpellodes above but near bottom of pistillode cavity, showing cavity (dotted) and nine vascular bundles consisting of two lateral vascular bundles (arrowed) and seven median vascular bundles. – Fig. 2: transverse section of 12-carpellate pistil just above ovule chamber, showing three lateral vascular bundles (arrowed) and twelve hairy ovuleless zones (dotted) separated by twelve locules each facing a median vascular bundle. – Fig. 3: transverse section of same pistil at middle level of ovule chamber, showing hairy ovule-bearing placentas (dotted), lateral vascular bundles opposite placentas and median vascular bundles opposite locules (the arrowed vascular bundles correspond to the two lower arrowed vascular bundles in Fig. 2). – Fig. 4: transverse section of same flower just beneath pistil cavity, showing



vascular bundles of pistil in central part and those of five associate staminodes at the periphery each with centripetal xylem (black) and centrifugal phloem (blank) (the arrowed vascular bundles correspond to the two lower arrowed vascular bundles in Fig. 2; the staminodes are similar to the right-arrowed staminodes in Fig. 7). – Fig. 5: transverse section of 3-armed partition between three carpels at about upper quarter of pistil, above ovule chamber, showing Y-shaped central parenchyma bordered at either side by 2–4-layered dotted sclerenchyma. – Fig. 6: transverse section of intercarpellary partition of same pistil above but near ovule chamber, showing central parenchyma with elongated cells (middle part).

since it is closest to it; the right staminode may be with the lower pistil for the same reason. It now has to be established with which pistil the middle staminode (left-arrowed) is associated. Although this staminode is further away from the upper pistil than from the other two pistils, the "one whorl criterion" suggests that it must be associated with the former pistil. In fact, if it were associated with either of the other two pistils, it would be in a second staminode-whorl around it. This contradicts the criterion. Therefore it must be with the upper pistil, for in this case it will be in the single staminode-whorl around that pistil. Further observation of the centripetalism of the xylem in the three staminodes at the bases of the flowers fully corroborated the relationship assumed for these staminodes.

Another essential criterion used is the "organic linking criterion". This was described briefly in Huynh (1991: 315) as being the "third point" for ascertaining the association of male organs in conjunction with the "centripetalism criterion". It is based on the fact that in angiosperms the male organs are generally fused along their basal part to the female organ of that flower, not to any other. As a result, in the transverse section at the uppermost level of this fusion the former organs appear partially fused to the latter organ (Fig. 7 and 10: for Fig. 7, consider the staminode just above and slightly left to the left-arrowed staminode). They are said to be "organically linked" to it. When such a section is observed, this linking (or partial fusion) may indicate that both organs are part of the same flower. The extent of the fusion varies from one species to another in *Freycinetia*. For example, the fusion of staminodes to pistil terminates more or less at the same level as the bottom of the pistil cavity in *F. banksii*, but clearly above this in *F. arborea* Gaudich. In fact, in the flower of *Freycinetia* the vascular bundles of the sex organs run both upwards and sideways from a common stock at the base (see Huynh 1991: Fig. 2 and 3). As a consequence, when the flower is observed in serial transverse sections, the vascular bundles of the male organs become distinct only from about the level of the flower base, where they are located at the periphery and may form protrusions, but they are still indistinct (Fig. 4 and 9). At subsequent upper levels they begin to become distinct but are still very close to the female organ while less so to nearby female organ(s), as shown by the two staminodes in the lower-right quarter of Fig. 7 and 8. It is at that level that "organic linking" may be observed (see above). At upper levels, the male organs become completely separated both from the female organ and the nearby female organ(s), as seen in the three distinct staminodes in the upper-right quarter of Fig. 7. At these levels, their association may be indicated by the centripetalism of their xylem but without certainty. In any case, whenever such problems are encountered, the solution consists in tracing the vascular bundles of the male organs down to the flower bases and in establishing which base(s) the vascular bundles originate from.

The above-mentioned criteria seem to allow to establish the male associates in any flower of *Freycinetia*. Application of them in *F. banksii*, however, required additional caution, as described in the next paragraph. The method can be tentatively applied to other species with large numbers of stigmas (e.g. *F. baueriana*; other species in Warburg 1900). In fact, as mentioned above, the flower of *F. banksii* is generally very wide. As a result, transverse sections are very long and comprise a large number of male components and locules, each of which corresponds to the middle part of a female component (carpel/carpellode). Consequently, they cannot be viewed under the microscope all in one field of view although a whole view of them was previously obtained with a stereo-microscope, and slides need to be frequently moved. In addition, with such a wide flower it is hardly possible to have all the vascular bundles of the male organs in one and the same transverse section at the basal level, this level being essential for recognition of the male associates in the flower of *Freycinetia* (see above). It follows that the number of male components cannot be ascertained other than by reliable restitution from two, three, or more consecutive sections.

The following method was devised to cope with these problems. It consists of four main steps. In the first step, the flower should be observed in transverse section a little above the bottom of the

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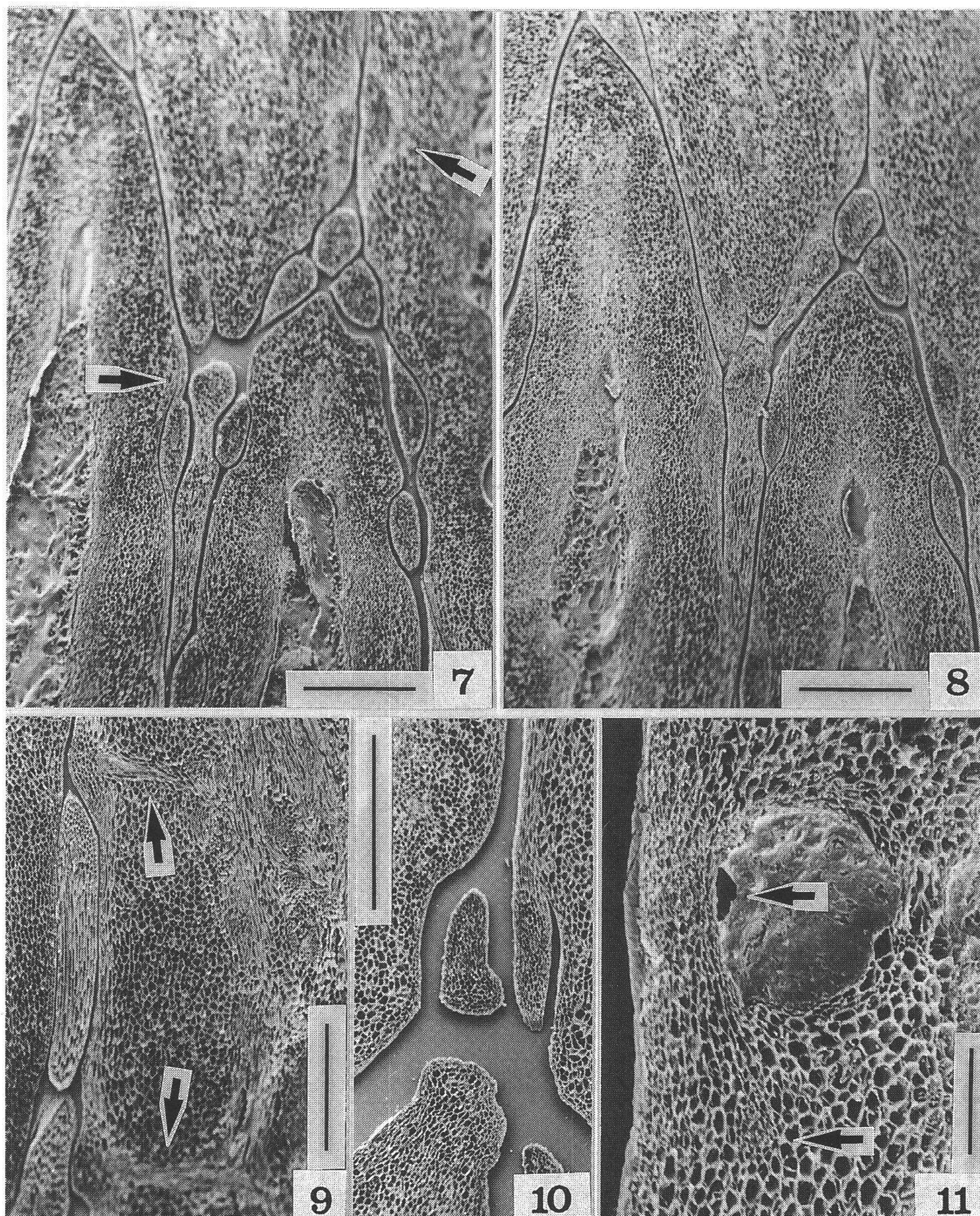
Fig. 7–11. *Freycinetia banksii* (features of pistillate spike at anthesis; spike axis vertical in all figures) (SEM). – Fig. 7: partial basal transverse sections of four flowers, showing staminodes [a staminode of upper flower left-arrowed; just above but slightly to left, another staminode; just to right of this, proximal end of pistil of that flower; of seven staminodes from above in right half



of figure, uppermost two (right arrow) are just perceptible, the subsequent belongs to upper flower, the left of two subsequent also to upper flower the right to lower flower, the subsequent to right flower, the lowermost to lower flower] (scale bar = 0.5 mm). – Fig. 8: partial transverse sections of same flowers but at a little lower level (compare staminodes with those in Fig. 7; scale bar = 0.5 mm). – Fig. 9: partial basal transverse section of flower, showing vascular strands (arrows) from vasculature of its pistil (whitish mass on right of arrows) to two protruding but indistinct staminodes on left (scale bar = 0.5 mm). – Fig. 10: partial basal transverse sections of three flowers, showing two staminodes in central part, left staminode (in centre) associated with left pistil, right staminode "organically linked" to right pistil (scale bar = 0.5 mm). – Fig. 11: partial transverse section of pistil just above ovule chamber, showing locule, lateral vascular bundle (lower arrow), and median vascular bundle (upper arrow) at middle point of pistil wall on left of locule (compare with Fig. 2) (scale bar = 100 μm).

pistil/pistillode cavity. At that level transverse sections of flowers are usually entire and male components are always cut through filaments. This facilitates the observation of both the number of locules and the centripetalism of the xylem in the male organs. Then a sketch of the section of the female organ (at that level) with all the locules should be made, and these should be numbered to be used as reference marks for the next identification of the male associates. In the second step, critical observation of the orientation of the xylem in the male components close to and around the female organ should be made, and the presumed male associates should be located on the sketch by referring to the nearby locules. In the third step, the flower should be observed in consecutive sections from the base up to the sketch level; and by using the criteria mentioned above, the associates of these male components should be critically checked. At the level of the flower base, in particular, the association of the male components could be further checked by observing the vascular strands from the vascular bundles of the male components to the central part of the base (Fig. 9). The strands are clearly visible in both flowers but more prominent in pistillate flowers because these have larger bases. In the fourth and final step, the number of male organs per flower is deduced. Thus, it requires much time and patience to study the numerical structure of the flowers in such species as *F. banksii*.

Results and discussion

1. Structure of the pistillate flower of Freycinetia banksii

This study involved determining the carpel number, then the staminode number, and noting other anatomical features of the flower.

The carpel number may be established by counting the stigmas. This was done when selecting flowers for sectioning. On some flowers, however, some stigmas were so close together that the counting was not reliable. Consequently, the carpel number in the flowers whose numerical structure was studied was only based on the placental number observed in transverse sections passing through various levels of the ovule chamber. In such sections (Fig. 3), each placenta could be readily recognized by the ovules and the hairs on it, and also by the two ovuleless and hairless locules flanking the placenta at either side. Probably this recognition was possible because the pistillate spikes studied were at an early fruiting stage. Indeed, in *F. arborea* most placentas could not be distinguished in more developed fruiting spikes, and establishing carpel numbers in these has been based on locule numbers in transverse sections above the ovule chambers. Recognition of placentas and locules was further facilitated by the fact that in the pistil wall, each locule was opposite a single vascular bundle – this was the median vascular bundle of the carpel corresponding to the locule – while each placenta faced several vascular bundles which were lateral vascular bundles, passing to placentas and generally not observed above ovule chambers except in a few cases (Fig. 2) (see below).

2–18 stigmas were counted on the fruiting spikes. Some pistils had more stigmas, up to about thirty, but they resulted from coalescence of two pistils since they showed two distinct ovule chambers. This feature seems to be a specific character, as pistil coalescence was observed in some species (e.g. *F. reineckei*) but not in others (e.g. *F. cumingiana*).

Thirty pistillate flowers were studied and all were single flowers (Table 1). No flowers with 15 and 17 carpels were found in the spikes studied.

Some noticeable anatomical features were observed in the pistil. One of these was that some, not all, of the lateral vascular bundles were found above the ovule chamber and extended up to beneath the apex of the pistil, as did the median vascular bundles. A small difference between them and the median vascular bundles was that the latter sometimes divided in the upper part of the pistil while the former apparently never did. These lateral

Table 1. Pistillate flowers studied

Female components	Male components	Female components compared with male components	Flowers studied
2 carpels	1 staminode	(+1)	1
2 carpels	4 staminodes	(−2)	1
2 carpels	6 staminodes	(−4)	1
3 carpels	7 staminodes	(−4)	1
3 carpels	8 staminodes	(−5)	1*
4 carpels	6 staminodes	(−2)	2
5 carpels	8 staminodes	(−3)	1
6 carpels	10 staminodes	(−4)	1
7 carpels	8 staminodes	(−1)	1
7 carpels	9 staminodes	(−2)	1
8 carpels	8 staminodes	(0)	1
8 carpels	10 staminodes	(−2)	1*
9 carpels	10 staminodes	(−1)	2
10 carpels	11 staminodes	(−1)	2
10 carpels	12 staminodes	(−2)	1
11 carpels	12 staminodes	(−1)	1*
12 carpels	10 staminodes	(+2)	1
12 carpels	12 staminodes	(0)	1
12 carpels	13 staminodes	(−1)	1
12 carpels	14 staminodes	(−2)	2*
13 carpels	14 staminodes	(−1)	2*
14 carpels	12 staminodes	(+2)	1
16 carpels	16 staminodes	(0)	1
18 carpels	15 staminodes	(+3)	2

* Two staminodes coalesced in the/one flower.

vascular bundles corresponded to some side-facing lateral vascular bundles at the placenta level. Consider, for example, the 12-carpellate pistil in Fig. 2–4. In the transverse section just above the ovule chamber, there are twelve semicircular locules and twelve hairy zones (Fig. 2). Each locule corresponds to a larger locule in the ovule chamber and each hairy zone to a hairy and ovule-bearing placenta (Fig. 3). Three locules each faced a pair of vascular bundles in the pistil wall, while the others each faced a single vascular bundle. No other vascular bundles were visible in the pistil. It had to be established if such a pair of vascular bundles comprised two median vascular bundles, or, instead, one median vascular bundle and one lateral vascular bundle. By tracing the pairs of vascular bundles down to the base of the pistil, it became evident that each pair comprised a median vascular bundle and a lateral vascular bundle. In fact, at the placental level (Fig. 3) the vascular bundles of the pistil were located on two more or less distinct circles: an outer circle, with twelve vascular bundles, each opposite the central part of a locule, and an inner circle, with twelve groups of vascular bundles, each group opposite a placenta. At the flower base (Fig. 4), the vascular bundles of the pistil were found on two more or less distinct circles too: the outer circle with twelve vascular bundles and the inner circle with twelve linear-shaped groups of smaller vascular bundles, each group being located between two consecutive vascular bundles in the outer circle. When tracing the vascular

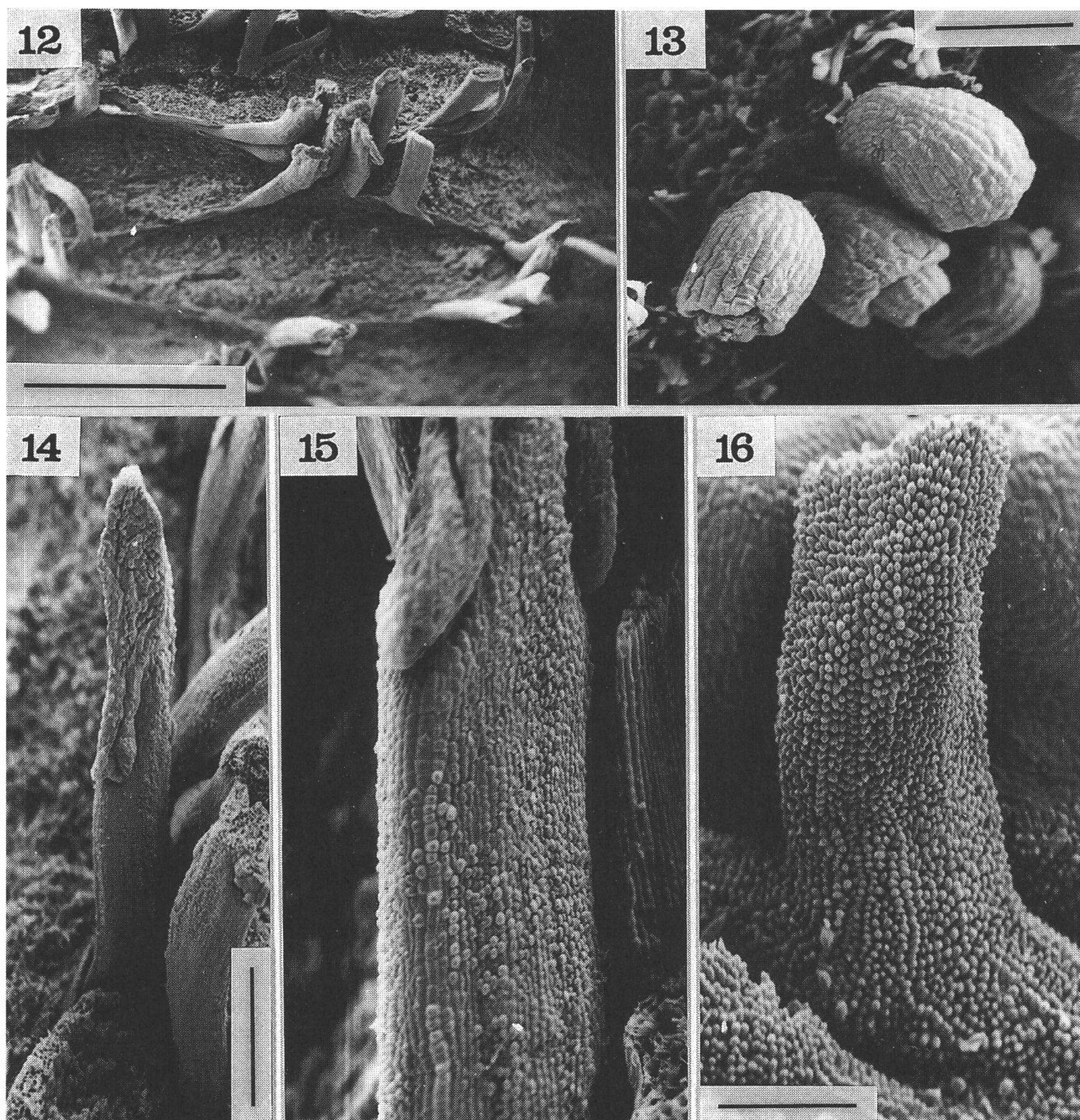


Fig. 12–16. *Freycinetia banksii* (SEM). – Fig. 12: pistillate spike at anthesis with pistils and staminodes removed, showing merging zone of four pistils (this is at the group of several staminode filaments; basal boundary of lower pistil entirely visible; spike axis horizontal in figure) (scale bar = 2 mm). – Fig. 13: ovules of young fruiting pistil (scale bar = 150 µm). – Fig. 14: staminode on pistillate spike at anthesis, showing anther and filament (scale bar = 0.5 mm). – Fig. 15: staminode filament on same spike (same magnification as Fig. 16). – Fig. 16: stamen filament on staminate spike at anthesis (scale bar = 150 µm).

bundles from that level back to above the ovule chamber, each vascular bundle in the outer circle was found in the central part of a locule, while each group of vascular bundles in the inner circle faced a placenta. Consequently, each former vascular bundle was a median vascular bundle and each latter vascular bundle a lateral vascular bundle. The tracing also revealed that the lateral vascular bundle in such a pair of vascular bundles corresponded to a side-facing vascular bundle in a linear-shaped group of the inner circle at the flower base and to a side-facing lateral vascular bundle at the placental level.

Another noteworthy anatomical feature was the central parenchyma situated from above the ovule chamber up to a little below the pistil apex. This parenchyma formed the central part of the intercarpellary partitions above the chamber. For example, in the transverse section at about 2 mm from the apex of a pistil about 5.2 mm long, the parenchyma showed two layers of more or less isodiametric cells and was bordered at either side by a 2-4-layered sclerenchyma (Fig. 5). At an upper level, where the carpel apices were distinct, the parenchyma was replaced by two distinct lignified one-layered epidermises. In the short lower part above the ovule chamber, the parenchyma showed 2 or 3 layers of more or less elongated cells, while each sclerenchyma zone was replaced by a parenchyma tissue generally formed of two layers of small and narrow cells (Fig. 6). In other species [e.g. *F. cumingiana*, *F. funicularis* (Savigny ex Lam.) Merr.], on the contrary, both the parenchyma and the two sclerenchyma zones bordering it are replaced by a single sclerenchyma (Huynh 1991: Fig. 8) which has many more cell layers than these.

Another specific anatomical feature of the pistil of *F. banksii* was its outer epidermis, in which crystal cells (each with one crystal) were of low density above the ovule chamber and lacking in the lower part. This contrasts with such species as *F. wilderi* Martelli where these crystal cells are abundant in both parts.

Such distinctive characters of *F. banksii* strongly suggest that pistil anatomy should be thoroughly investigated in further taxonomic revisions in *Freycinetia*, for tentatively distinguishing between close species, e.g. *F. banksii* and *F. baueriana* (see above). In the small number of species in which pistil anatomy has been studied, each can be readily identified by this anatomy, disregarding the well-known character of carpel numbers.

In the fruiting pistils narrow fibre-strands with crystal cells were observed in the middle part. Other lignified cells (either isolated or in small groups) but without crystal cells were also found in that part. Narrow strands of crystal cells without lignified cells were visible between the fibre strands zone and the ovule chamber. All three features, however, were not observed in the pistils at anthesis. Also noteworthy is the fact that mucilage in the pistil was so abundant that it was frequently extruded onto stigmas, a feature not observed in other species.

In the staminodes the anther was more or less as long as the filament, sometimes distinctly shorter, and generally a little wider. But rod-shaped staminodes with anther as wide as filament (Fig. 14) were frequently observed. The anther showed no endothecial thickenings when squashed in Phloroglucinol-HCl stain, and the filament was both smooth and verrucate (Fig. 15). Some staminodes coalesced along their filaments (see Discussion and conclusion).

2. *Structure of the staminate flower of Freycinetia banksii*

This study involved determining the carpellode and stamen numbers, and noting other anatomical features of the flower.

All the staminate flowers whose numerical structure could be satisfactorily investigated were systematically studied and entered in the accounts. In addition, in all the species of *Freycinetia* studied with both pistillate and staminate material, the same species showed the same numbers of stamens and staminodes and the same numbers of carpellodes and carpels. This perfect floral homology seemed to be corroborated by *F. banksii* as well (see below). Considering both this homology and this statistical aspect of the study, some features from the numerical structure of the staminate flower of this species (for example, the most frequent numbers of male and female components) may be

Table 2. Staminate flowers studied

Female components	Male components	Female components compared with male components	Flowers studied
4 carpellodes	5 stamens	(−1)	1
4 carpellodes	6 stamens	(−2)	1*
4 carpellodes	7 stamens	(−3)	1
5 carpellodes	7 stamens	(−2)	1
5 carpellodes	8 stamens	(−3)	1
6 carpellodes	7 stamens	(−1)	2
6 carpellodes	8 stamens	(−2)	5
6 carpellodes	9 stamens	(−3)	1*
7 carpellodes	7 stamens	(0)	3
7 carpellodes	8 stamens	(−1)	3
7 carpellodes	9 stamens	(−2)	2
7 carpellodes	10 stamens	(−3)	2
7 carpellodes	11 stamens	(−4)	3*
8 carpellodes	7 stamens	(+1)	1
8 carpellodes	9 stamens	(−1)	3
8 carpellodes	10 stamens	(−2)	2*
8 carpellodes	11 stamens	(−3)	2**
8 carpellodes	12 stamens	(−4)	1***
9 carpellodes	8 stamens	(+1)	1
9 carpellodes	9 stamens	(0)	2
9 carpellodes	10 stamens	(−1)	1
9 carpellodes	12 stamens	(−3)	1*
10 carpellodes	10 stamens	(0)	1
11 carpellodes	13 stamens	(−2)	2*
12 carpellodes	10 stamens	(+2)	1
12 carpellodes	11 stamens	(+1)	1
13 carpellodes	11 stamens	(+2)	1

* Two stamens coalesced in the/one flower.
** Two stamens coalesced in each flower.
*** Two pairs of coalesced stamens.

reasonably extended to the pistillate flower. Consequently, it was sufficient and advantageous to use staminate material, instead of pistillate material or both, for a statistical study of the numerical structure of the flower of this species since much fewer preparations were needed, staminate flowers being arranged much more densely than pistillate flowers. It was found unnecessary to make a similar statistical study with pistillate material for corroboration. Indeed, some corroboration was already obtained (see Tables 1 and 2: for example, the 7-carpellate flowers with 8 and 9 staminodes correspond well to the staminate flowers with 7 carpellodes which have 8 and 9 stamens); and given the wide range of variation in the numerical structure shown by both flowers, it appeared hardly possible to find pistillate flowers corresponding to all of the staminate flowers observed, supposing a statistical study of the flower structure with pistillate material was also made.

The numerical structure in forty six single staminate flowers was studied (Table 2). The carpellode numbers were found by counting the pistillode locules. Transverse sec-

tions of pistillodes in this species were elongated and showed a certain number of more or less semicircular locules along both sides and two narrow locules at the apices (Fig. 1 and 17), "lateral locules" (Fig. 18) and "apical locules", respectively. Each in its central part was opposite a vascular bundle, which is a median vascular bundle as established in other species (Huynh 1991: 301). The lateral locules were the only sites of abundant mucilage in the pistillode.

Vascular bundles were also observed in some, not all, of the interlocular zones of the pistillode wall (Fig. 1: arrowed vascular bundles). They were generally much shorter than the median vascular bundles. This feature and the interlocular position readily distinguished them from these. The observation of lateral vascular bundles in the pistillode of *F. banksii* was unexpected since no lateral vascular bundles were found in the pistillodes of the other species studied (e.g. *F. cumingiana*, *F. funicularis*, *F. javanica* var. *expansa*, *F. reineckei*, *F. scandens*). In any case, whether they are true lateral vascular bundles as assumed in the present paper, or, instead, additional median vascular bundles, the unusual vascular bundles in interlocular zones of the pistillode wall of *F. banksii* apparently isolate this species in *Freycinetia*. It would be of interest to see if this unusual feature exists in other species too or has some taxonomic significance at the sectional level. In this respect, *F. banksii* belongs in Sect. *Freycinetia* (Stone 1968: 368) and was the first species of the section to be studied as in the present paper. Also noticeable is the fact that this pistillode has two kinds of locules (Fig. 1): the lateral locules, wide and more or less semicircular; the apical locules, narrow and generally linear.

The pistillodes of *F. banksii* varied considerably in size. Generally speaking, those with high carpellode-numbers were larger than those with low numbers (Fig. 19). Variable sizes were observed among pistillodes with the same carpellode-numbers as well. No correlation was apparent between this variation and stamen numbers, as observed in the five flowers with 7 carpellodes in Table 2 studied for this purpose: 7 stamens, transverse sections of pistillode at middle level about 2230 μm in longer diameter; 8 stamens, about 1923 μm; 9 stamens, about 2300 μm; 10 stamens, about 2000 μm; 11 stamens, about 2076 μm.

Furthermore, no lignified cells were observed in the pistillode except for the xylem of the vascular bundles, and no partition walls were visible between the carpellodes as also observed in the other species studied. This seems to indicate that the pistillode of *Freycinetia* corresponds to the ovule chamber of the pistil since this latter has partition walls in the upper part but not in the ovule chamber. As a result, absence vs presence of partition walls provides another distinctive character between these two female organs (other characters were described in Huynh 1991). This distinction, however, is only valid for adult organs because young pistils do not have any partition walls either, as recently observed in *F. funicularis* by Huynh [The flower structure in the genus *Freycinetia*, Pandanaceae (part 2) – Early differentiation of the sex organs, especially of the staminodes, and further notes on the anthers – in press in Bot. Jahrb. Syst.]. This distinctive feature of pistillode from pistil was also shown by *Pandanus* sect. *Martellidendron*, with 2-carpellate pistils (Huynh 1981: 48 and Fig. 20–22).

The staminal features of *F. banksii* observed are as follows. The anthers belonged to the *Funicularis* type, characterized by anthers generally more or less cordiform, with an apical notch, and by adaxial and abaxial pollen-sacs both converging at the apex without coalescing (Huynh, in press in Bot. Jahrb. Syst.: see above). The endothecium comprised one cell-layer. The extent of the endothecial thickenings was more or less the same as in *F. cumingiana* (Huynh 1991: Fig. 5). No endothecial thickenings were observed in the generally distinct epidermal cell layer, contrary to *F. funicularis* where "endothecial bands

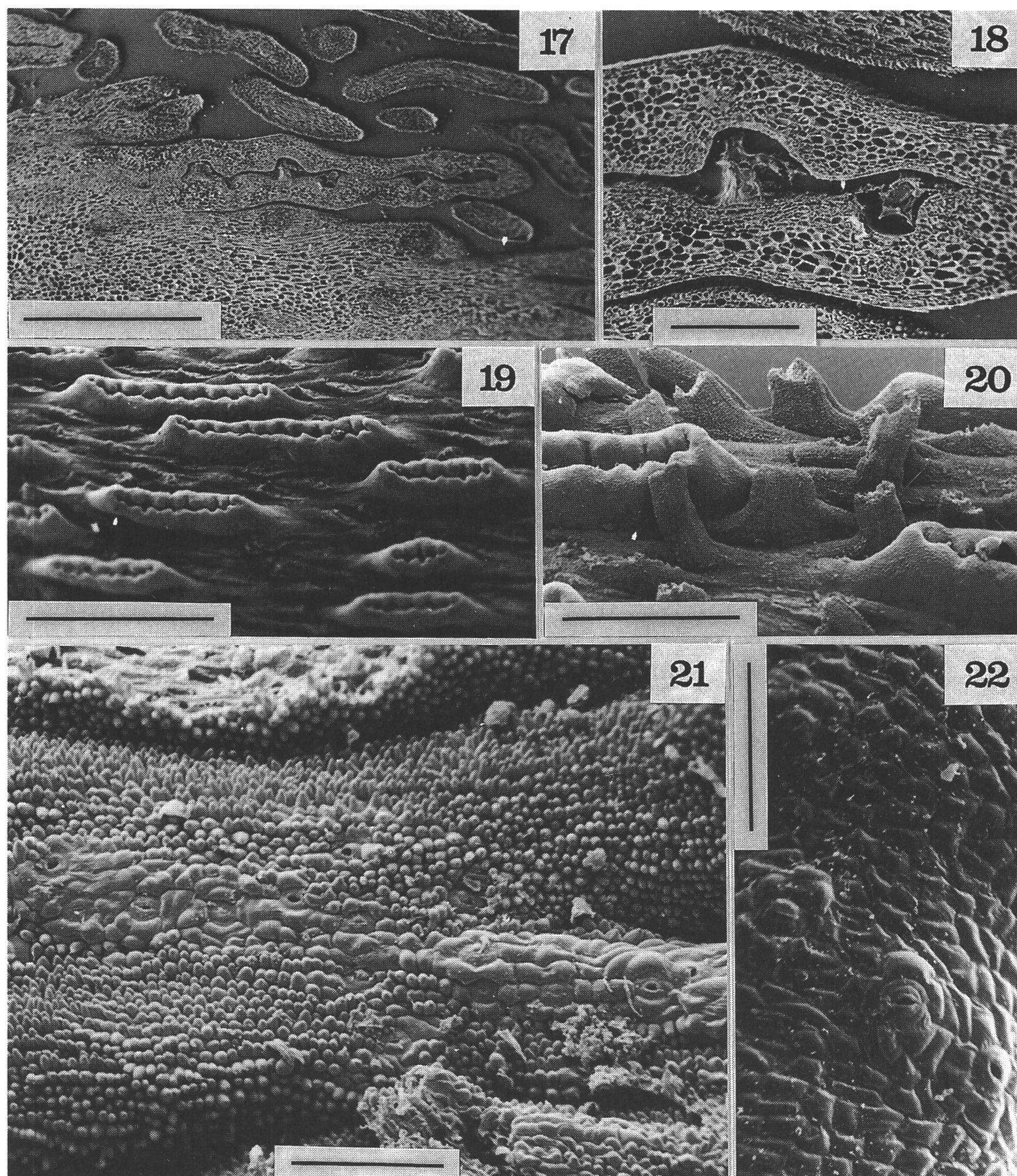


Fig. 17–22. *Freycinetia banksii* (features of staminate spike at anthesis; spike axis horizontal in Fig. 17–21) (SEM). – Fig. 17: middle transverse section of pistillode (middle zone) and filament sections (above and on right) (scale bar = 1 mm). – Fig. 18: central part of pistillode section in Fig. 17, showing two lateral locules with mucilage (scale bar = 0.2 mm). – Fig. 19: spike with stamens removed, showing variable pistillodes (scale bar = 2 mm). – Fig. 20: other part of same spike, showing wide filament (in centre) resulting from coalescence of two normal filaments (scale bar = 1 mm). – Fig. 21: surface of same spike, formed of smooth/verrucate zone (middle part) and papillate zones (scale bar = 150 µm). – Fig. 22: outer face of pistillode wall, with smooth surface and stomata (scale bar = 100 µm).

are formed in the epidermal cells instead of . . . the subepidermal cells" (Ganguly 1976: 271). The vascular bundle extended up to about the upper quarter of the anther. The pollen was smooth. The filaments were papillate (Fig. 16). The spike surface between stamen filaments was smooth and verrucate in some sites but generally papillate (Fig. 21). The collenchyma in the filaments was clearly visible, with thick walls. It extended up to a little above the filament apex and down to a little beneath the spike surface, at which level it slightly expanded laterally. Furthermore, some stamens coalesced (see below).

Discussion and conclusion

The numerical structure of the flower of *F. banksii* may be deduced from the data in Tables 1 and 2. These show a very large number of flower types in both sexes, but we are fairly sure that other types also exist. In pistillate flowers the carpels ranged from 2 to 18, the staminodes from 1 to 16. Flowers with 2 or 3 carpels, however, were rare. In staminate flowers the carpellodes ranged from 4 to 13, the stamens from 5 to 13. No staminate flowers with 2 or 3 or 14–18 carpellodes or with 1–4 or 14–16 stamens were found, but such flowers may have been observed if a much larger number of staminate flowers had been studied. Furthermore, the numbers of male components varied in an unusual way in some flowers (for example, 1, 4 and 6 staminodes in the three 2-carpellate flowers studied: see Table 1). A suitable explanation remains to be found. It would be of interest to know if similar variations exist in other species with high stigma-numbers (e.g. *F. baueriana*).

Both flowers, however, showed some important common features, which may be considered the floral characters of *F. banksii*. In general, there are more male components per flower than female. Basically, the staminode number is the same as the stamen number and the carpellode number the same as the carpel number, but there is much variation. In addition, since the numerical structure was studied statistically in the staminate flowers, and considering the perfect floral homology also observed in the other species studied with both pistillate and staminate material (see above), it may be assumed that the most frequent numbers of male components in this species range from 7 to 11, and female components from 6 to 9. In a previous paper, Huynh (1991: 326) suggested that in *Freycinetia* a correlation seems to exist between the carpel number and the stamen number, and that the staminate flower may not in general have more than about twenty stamens. The data from the flower of *F. banksii* confirm the existence of such a correlation in this species, but reveal that this is not a simple correlation. They also indicate that the highest number of stamens in *Freycinetia* (see Introduction) may not even be about twenty, but probably about fifteen, since 16 staminodes were the highest number of male components observed in this species.

Furthermore, in *F. banksii* two nearby stamens or staminodes were coalesced along their filaments in some flowers (Fig. 20). Tracing their vascular bundles down to the flower bases revealed that these filaments belonged to the same flower. In such a flower, the coalesced filaments were found at an end of the transverse sections of the female organ. This may be explained, since each end of the sections was generally located in the merging zone of three or four flowers (Fig. 7 and 12). As a result, filaments were denser there than elsewhere, which might increase the chances of filaments to coalesce. Despite the strong congestion of flowers, no coalescence was observed involving two filaments of two different flowers.

These common features of the pistillate and the staminate flower in conjunction with those features mentioned above revealed the structural similarity of these flowers, confirming the perfect floral homology observed in other species. Other common features were the elongated shape of both the pistil and the pistillode, both being similarly oriented along the spike axis, the abundance of mucilage in both of them, the cavity hairs sometimes observed in the pistillode, and, in particular, the lateral vascular bundles in the pistillode which corresponded to those in the pistil.

These structural similarities and the perfect homology between the pistillate and the staminate flower revealed the potential bisexuality of this species. The potential bisexuality of *Freycinetia* (Huynh 1991: 318) has a very variable expression. For example, two female plants with bisexual spikes but without hermaphrodite flowers were exceptionally observed in the populations of *F. banksii* in the Paparoa National Park (Lord 1991: 85); on the contrary, bisexuality was found to be common in the Upolu populations of *F. reineckei* (Cox 1981: 197), and hermaphrodite flowers were differentiated in some of its bisexual spikes as well (Huynh & Cox, in press in Bot. J. Linn. Soc.: see above). It may be of interest to investigate, at least as a working hypothesis, if the rarity of sex change in the former species may to some extent be correlated to the exceptionally wide variation in the numerical structure of its flower. Hermaphrodite flowers were also observed in bisexual spikes of *F. cumingiana* (Huynh 1991: 308), where the same individuals may occasionally produce either pistillate or staminate inflorescences, or both, and sometimes mixed inflorescences, formed of pistillate spike(s) and either staminate spike(s) or bisexual spike(s) (Poppendieck 1987: 315 and Fig. 2; Huynh 1991: 308 and 318). Other cases and forms of occasional monoecism have been observed in *F. imbricata* Blume var. *hispidula* Stone (Stone 1972: 207), *F. negrosensis* Merr. (Stone, as quoted in Cox 1981: 195), *F. scandens* (Cox et al. 1984), *F. funicularis* (Cox et al. 1984: 313; Poppendieck 1987), and *F. scabripes* Warb. (Stone 1990: 43).

References

Cox P. A. 1981. Bisexuality in the Pandanaceae: new findings in the genus *Freycinetia*. Biotropica 13: 195–198.

Cox P. A., Wallace B. & Baker I. 1984. Monoecism in the genus *Freycinetia* (Pandanaceae). Biotropica 16: 313–314.

Ganguly B. 1976. Some observations on the flower of *Freycinetia funicularis*. Sci. & Cult. 42: 270–272.

Huynh K.-L. 1981. *Pandanus kariangensis* (sect. *Martellidendron*), une espèce nouvelle de Madagascar. Bull. Mus. Natn. Hist. Nat., Paris, 4e sér., 3, sect. B, Adansonia 1: 37–55.

Huynh K.-L. 1991. The flower structure in the genus *Freycinetia*, Pandanaceae (part 1) – Potential bisexuality in the genus *Freycinetia*. Bot. Jahrb. Syst. 112: 295–328.

Lord J. M. 1991. Pollination and seed dispersal in *Freycinetia baueriana*, a dioecious liane that has lost its bat pollinator. New Zealand J. Bot. 29: 83–86.

Moore L. B. & Edgar E. 1970. Flora of New Zealand, vol. 2. Government Printer. Wellington.

Moore L. B. & Irwin J. B. 1978. The Oxford book of New Zealand plants. Oxford University Press.

Poppendieck H.-H. 1987. Monoecy and sex changes in *Freycinetia* (Pandanaceae). Ann. Missouri Bot. Gard. 74: 314–320.

St. John H. 1973. Revision of the genus *Pandanus* Stickman (part 35): Additional *Pandanus* species from New Guinea. Pacific Sci. 27: 44–91.

Stone B. C. 1968. Materials for a monograph of *Freycinetia* Gaud. IV. Subdivision of the genus, with fifteen new sections. Blumea 16: 361–372.

Stone B. C. 1972. Materials for a monograph of *Freycinetia* Gaud. (Pandanaceae) XV. The Sumatran species. Federation Mus. J. n. s. 15 (for 1970): 203–207.
Stone B. C. 1973. Materials for a monograph of *Freycinetia* Gaudich. XIV. On the relation between *F. banksii* A. Cunn. of New Zealand and *F. baueriana* Endl. of Norfolk Island, with notes on the structure of the seeds. New Zealand J. Bot. 11: 241–246.
Stone B. C. 1990. New evidence for the reconciliation of floral organisation in Pandanaceae with normal angiosperm patterns. *In* P. Baas, K. Kalkman & R. Geesink (eds.), The Plant Diversity of Malesia: 33–55. Kluwer (Dordrecht, The Netherlands).
Warburg O. 1900. Pandanaceae. *In* A. Engler (ed.), Das Pflanzenreich 3 (=IV.9). Engelmann (Leipzig).