

Studies in the southern African species of *Justicia* and *Siphonoglossa* (Acanthaceae): palynology

K.L. IMMELMAN*

Keywords: Acanthaceae, *Justicia*, palynology, *Siphonoglossa*, southern Africa, taxonomy

ABSTRACT

The gross morphology of the pollen of all southern African species and subspecies of *Justicia* and *Siphonoglossa* was investigated, as well as that of eight tropical African species of *Justicia*. The following pollen types were found in the southern African species of *Justicia*: two- or three-colporate, each with the margocolpus either entire or broken up into areolae, and two-porate areolate pollen. One tropical African species had the colpus very short, and in another tropical African species it was replaced by an extra row of areolae. All *Siphonoglossa* species had two-colporate pollen with areolae and long colpi. The southern African species of *Justicia* could be separated from *Siphonoglossa* on pollen characters, and some sections of *Justicia* could also be distinguished on the same basis.

UITTREKSEL

Die algemene morfologie van die stuifmeel van alle spesies en subspesies van *Justicia* en *Siphonoglossa* in Suider-Afrika, asook dié van agt *Justicia*-spesies uit tropiese Afrika, is ondersoek. Die volgende stuifmeel-tipes is by die *Justicia*-spesies van Suider-Afrika gevind: twee- of driekolporaat, elk met die margokolpus of gaaf of opgebreek in areole, en tweeporate areolêre stuifmeel. By een spesie uit tropiese Afrika was die kolpus baie kort, en by 'n ander uit tropiese Afrika is dit deur 'n ekstra ry areole vervang. Alle *Siphonoglossa*-spesies het tweekolporate stuifmeel met areole en lang kolpusse gehad. Die *Justicia*-spesies van Suider-Afrika kon op grond van stuifmeelkenmerke van *Siphonoglossa* geskei word, en sommige *Justicia*-seksies kon ook op dieselfde basis onderskei word.

INTRODUCTION

The gross morphology of the pollen of all southern African species and subspecies of *Justicia* and *Siphonoglossa* was investigated, as well as that of eight tropical African species of *Justicia*. In order to place the studies into perspective, brief reference is made to findings of other researchers.

Radlkofer (1883) was the first to point out the potential value of the pollen in classification of the Acanthaceae, but it was Lindau (1895) who first attempted to use the pollen systematically in its classification.

The 'typical representatives' of the subtribe Justiciinae, according to Bremekamp (1965), have prolate, dorsiventrally flattened pollen, with the pore in the middle of a trema area which is studded with circular 'insulae' (areolae).

Stearn (1971), after discussing the pollen type of a number of Jamaican species of *Justicia*, concluded that 'The palynology of the group as a whole must certainly have a significant part in its reclassification. Pending that reclassification, *Justicia* is probably best accepted in a broad sense'.

Gibson (1972) widened the circumscription of *Justicia* and, if her delimitation is accepted, the range of pollen types present in the genus will be greatly increased, and will include 3- and 4-porate pollen and pollen with the areolae scattered across the face of the grain.

Balkwill & Getliffe Norris (1988), who reappraised the tribal and subtribal limits of Acanthaceae in southern Africa, considered that in southern Africa each tribe had a characteristic pattern of pollen 'against which aberrant genera can be contrasted'. They considered prolate tricolporate grains with entire margocolpi as being the primitive state.

Graham (1989) surveyed a number of species of *Justicia* and related genera worldwide, and greatly widened the circumscription of the genus to include a number of formerly segregate genera, including *Siphonoglossa*. She divided the genus into a number of sections and subsections which have been followed here, and considered pollen, in combination with other characters, to be of major taxonomic importance. She considered the South African species of *Siphonoglossa* to belong in *Justicia*, but not in the same section as the South American species of *Justicia*. She described 10 pollen types, but did not confirm the 4-aperturate pollen types reported for a few New World species.

METHODS AND MATERIALS

In this study the pollen of all southern African species and subspecies of *Justicia* and *Siphonoglossa* was investigated with the SEM. Taxa covered were all those occurring in southern Africa as defined by the *Flora of southern Africa*. Eight tropical African species were also investigated for purposes of comparison, but were not measured.

Material was obtained from three sources: herbarium sheets, fresh material grown in the greenhouse at the Botanical Research Institute, Pretoria, and material preserved in alcohol (gathered in the field).

* Botanical Research Institute, Department of Agriculture and Water Supply, Private Bag X101, Pretoria 0001.

MS. received: 1988.07.16.

Fresh material of two of the species, *J. flava* (Vahl) Vahl and *J. protracta* (Nees) T. Anders. subsp. *protracta*, was also acetolysed (following Erdtman 1971, as quoted by Taylor, Hollingsworth & Bigelow 1974), before being mounted and viewed. The acetolysed material was then compared with that of the same species mounted directly onto stubs.

One to four specimens of each species and subspecies of *Justicia* and *Siphonoglossa* in southern Africa were examined. The anthers were macerated onto 15 mm aluminium stubs covered with double-sided adhesive tape. Thin tape was used, as the pollen grains tended to sink partially into the thicker gum of some tapes and became difficult to photograph well.

After air-drying, the mounted material was glow-discharge-coated with $\pm 400 \text{ \AA}$ of metallic gold in an Eiko sputter coater. The specimens were then viewed with an MSM 4 Hitachi-Akashi (desk top model) SEM at kv 15. Selected views were photographed of the profile (face), side and amb (polar) faces of the pollen grains of each taxon, using a Mamiya 6×7 camera and Ilford FP4 125 ASA film. The film was developed in Microdol X.

The size of the pollen grains was measured from water-mounted pollen with a Kontron Image analyser.

The terminology used is illustrated in Figure 1.

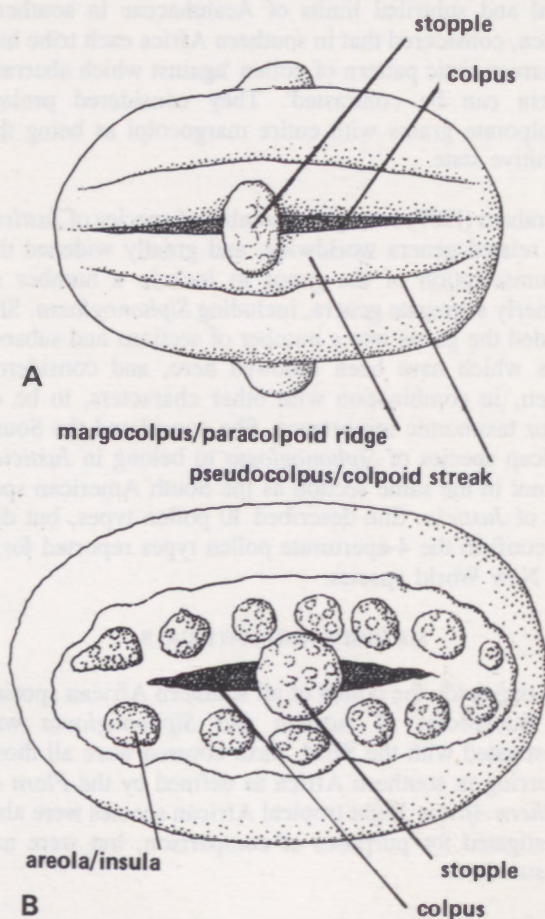


FIGURE 1.—Diagrams of pollen grains in *Justicia* and *Siphonoglossa*: A, with entire margocolpus, $\times 2400$; B, with areolae, $\times 2400$.

RESULTS AND DISCUSSION

No differences were observed between pollen taken from fresh and from dried material when it was viewed.

No significant difference was found between acetolysed material and material mounted directly onto the stubs. This is similar to the findings of Munday (1980) in *Monechma*, except that acetolysed material tended to lose its 'stopples' and had a high proportion of damaged grains. Material was therefore viewed throughout without pretreatment, and results given are of such pollen.

Shape of pollen grains

The pollen of both *Justicia* and *Siphonoglossa* was found to be basically prolate, with some species, e.g. *J. orchioides* L. f., *J. platysepala* (S. Moore) P.G. Mey. and *J. thymifolia* (Nees) T. Anders., showing a slight median constriction (Figure 5C).

Sexine pattern

The sexine pattern was lophate or reticulate over most of the grain with smooth areas (margocolpi) on either side of the colpi.

These margocolpi were ornamented either with an entire band of lophate sexine (paracolpoid ridge) (Figure 1A) or this band was broken up into areolae, i.e. circular areas of lophate sexine (Figure 1B). The areolae were in one or two rows (*Justicia*) (Figure 3A; 4D, G; 5A) or sometimes in up to three rows (*Siphonoglossa*). The outer row was frequently partially merged with the main area of lophate sexine. In one tropical African species, *J. goetzei* Lindau, the area normally occupied by the colpus was occupied instead by a single row of areolae, as well as the row on either side, i.e. a total of three rows of areolae in the pair of margocolpi.

In the case of pollen with entire paracolpoid ridges, the colpoid streak (which appears as a furrow between the paracolpoid ridge and the main area of lophate sexine) would sometimes continue to the poles. This, however, may be variable (see also Balkwill & Getliffe Norris 1985), and has not been used in the classification. The pseudocolpi may actually join at the poles, for instance in *J. campylostemon* and *J. glabra* among the three-colporate species (Figure 2D).

Pores and colpi

The pollen grains have either two (Figures 4 & 5) or three pores (Figures 2 & 3). Each may lie within an elongate colpus running to near each pole. It is sometimes difficult to establish with certainty whether there is a colpus or not. Two species, however, *J. anselliana* (Nees) T. Anders. and *J. anagalloides* (Nees) T. Anders., were definitely without colpi (Figure 4G). An intermediate stage was seen in one of the tropical species, *J. elegantula* S. Moore, where the colpi were present but short. Another tropical African species, *J. goetzei*, had a single row of areolae where the colpus would normally be. In all species a granular stopple was present projecting from each pore (Figure 4B, C).

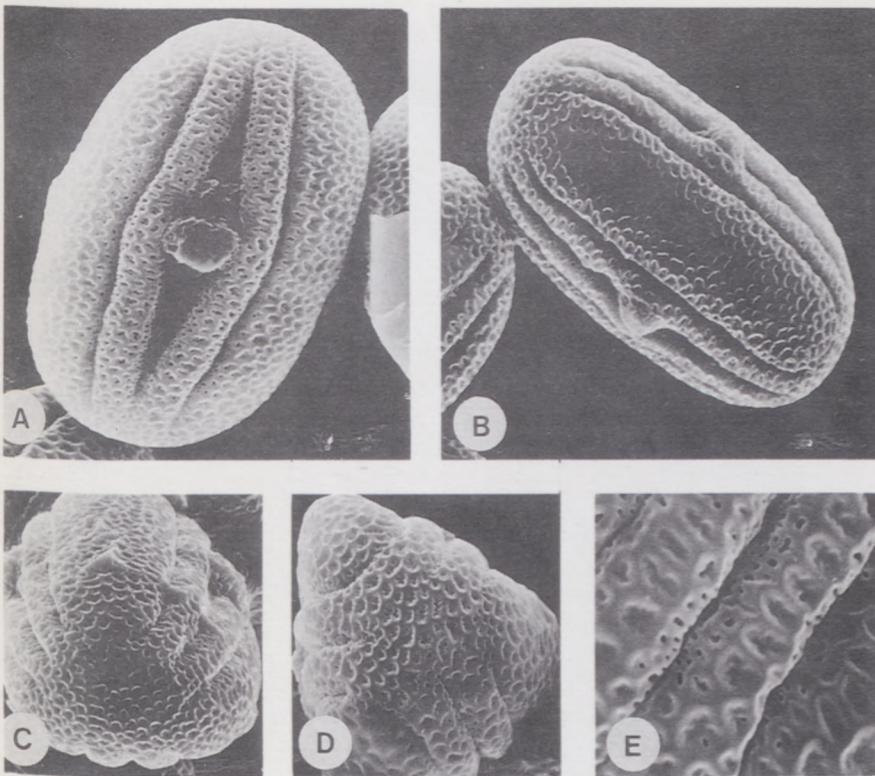


FIGURE 2.—Pollen of *Justicia* species, 3-colporate, margo-colpi entire. A–C, *J. betonica*, *Strey 8161*: A, face; B, side; C, amb, all $\times 1200$. D–E, *J. glabra*, *Smith 2734*: D, amb showing pseudocolpi continuous at poles, $\times 1200$; E, surface detail, $\times 2900$.

Length

The length of the pollen grains (taken between the poles) covered a wide range in the two genera. In *Justicia* it varied from 22 μm [*J. anselliana*] to 77 μm [*J. petiolaris* (Nees) T. Anders.] and in *Siphonoglossa* from 58 μm [*S. leptantha* (Nees) Immelman] to 90 μm [*S. linifolia* (Lindau) C.B. Cl.]. The pollen in the genera therefore varies from small to medium to large according to the categories of Erdtman (1952).

Size (length) of pollen grain, although used in the key below, has not been used to reach taxonomic decisions,

as there is a large range in pollen sizes in each species, even in measurements taken from a single flower. Keys using pollen sizes would need to be drawn up with caution, and be based on a number of pollen samples from different individuals and populations.

Abnormal grains

These are seen occasionally in both genera, and take the form of a much larger than average, triangular grains with a trilete colpus (Figure 5F). It is presumed that these are what Bhaduri (1944) called 'giant grains'.

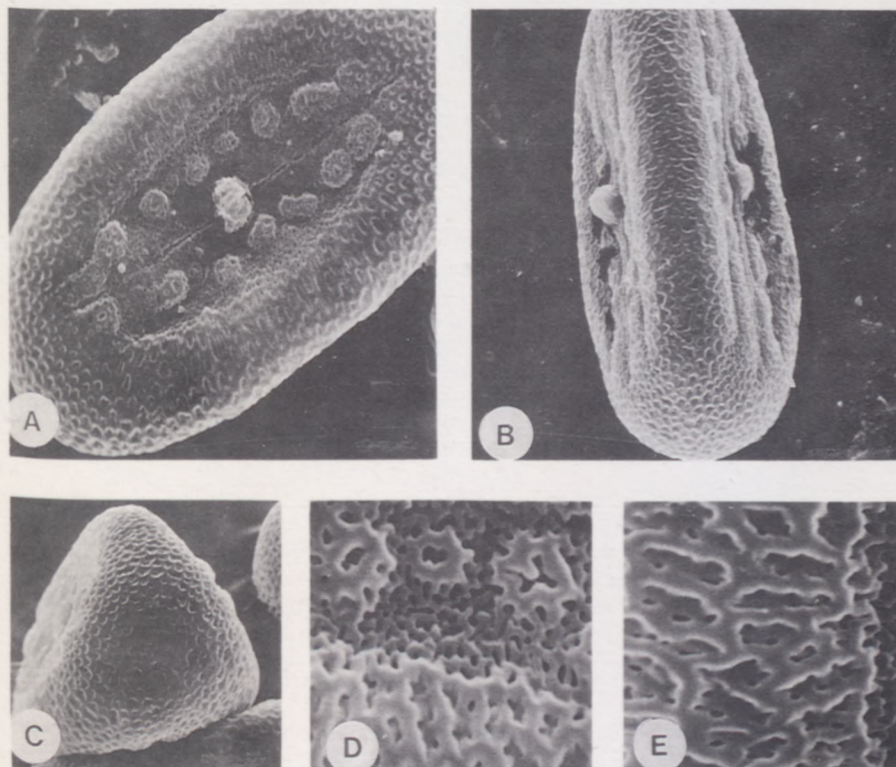


FIGURE 3.—Pollen of *Justicia* species, 3-colporate, margo-colpi areolate. A & C, *J. kirkiana*, *Muir 1090*: A, face, $\times 1400$; C, amb, $\times 10$. B, *J. petiolaris* subsp. *incerta*, *Meeuse 9100*, side, $\times 1000$. D & E, *J. flava*, *Werdermann & Oberdieck 1744*, surface detail showing areolae, $\times 4000$.

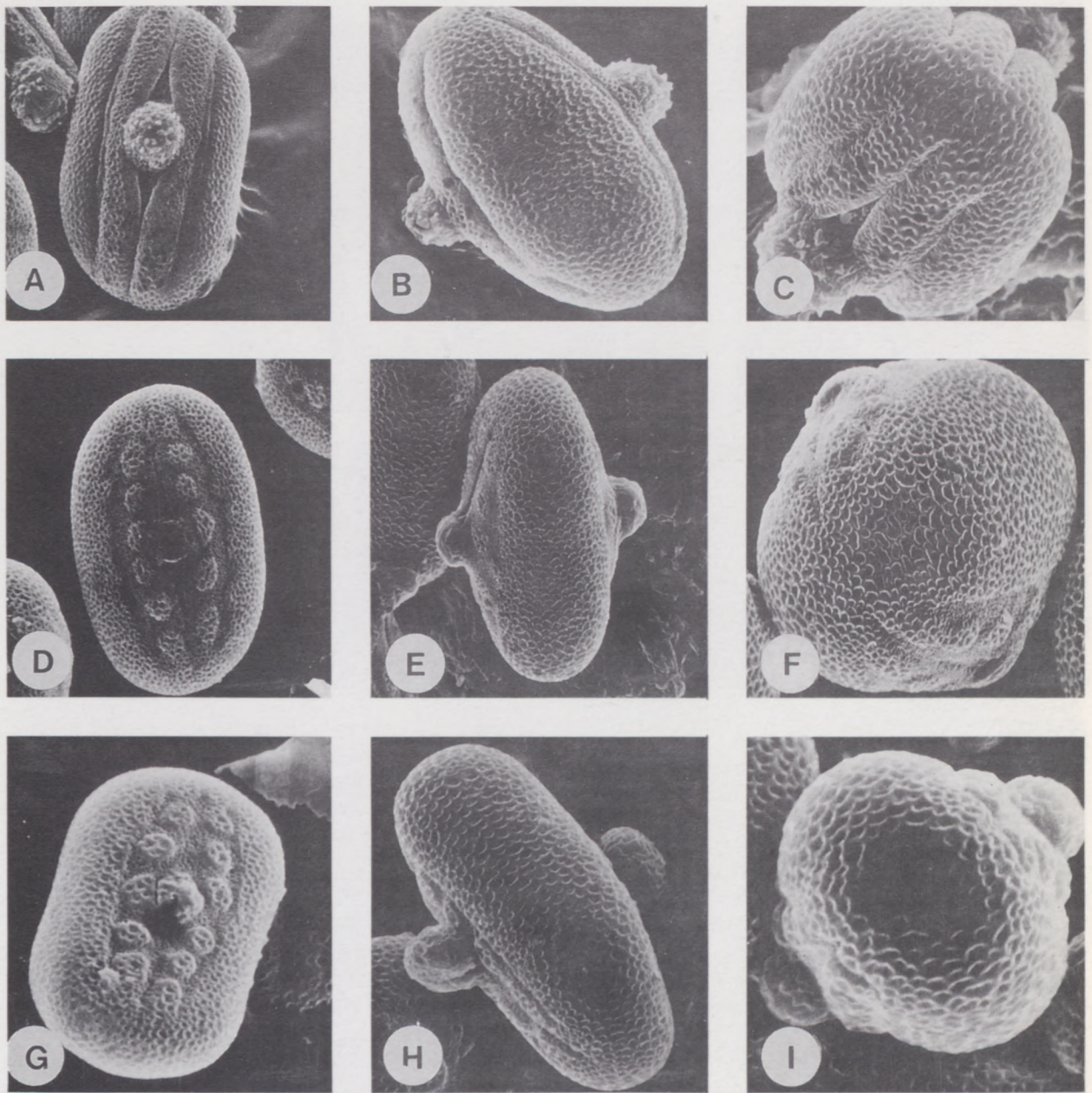


FIGURE 4.—Pollen of *Justicia* species, 2-colporate. Margocolpi entire: A–C, *J. guerkeana*, Barnard 122: A, face, $\times 1200$; B, side, $\times 1200$; C, amb, $\times 1600$. Margocolpi areolate, D–I. D–F, *J. capensis*, Acocks 20100: D, face, $\times 900$; E, side, $\times 900$; F, amb, $\times 1400$. G–H, *J. anagaloides*, Hafstrom & Acocks 1390: G, face, $\times 1800$; H, side, $\times 1800$. I, *J. anselliana*, De Winter 2518, face, $\times 3000$.

Description of pollen of species seen (number in brackets is the mean length, $n = 20$):

Justicia

- anagaloides* (Nees) T. Anders.: two-porate, areolate, one row areolae, length 28 (31,1) 36 μm (Hafstrom & Acocks 1390) (Figure 4G, H).
anselliana (Nees) T. Anders.: two-porate, areolate, one row areolae, length 22 (31,3) 29 μm (De Winter 2518) (Figure 4I).
betonica L.: three-colporate, margocolpus entire, length 36 (46) 58 μm (Strey 8161) (Figure 2A, B, C).
bolusii C.B. Cl.: three-colporate, margocolpus entire, length 34 (46) 58 μm (Immelman 376).
campylostemon (Nees) T. Anders.: three-colporate, margocolpus entire, length 45 (49,8) 53 μm (McClellan 126).
capensis Thunb.: two-colporate, areolate, one row areolae, length 48 (51,2) 60 μm (Acocks 20100) (Figure 4D, E, F).
caerulea Forssk.: two-colporate, areolate, one row areolae, length not measured (Polhill & Paulo 1022).
cordata (Nees) T. Anders.: two-colporate, margocolpus entire, length not measured (Greenway & Kanuri 11357).

- crassiradix* Burkill & C.B. Cl.: two-porate, areolate, one row areolae, length unknown (Killick & Leistner 3417).
cuneata Vahl (all three subsp.): two-colporate, margocolpus entire, length (subsp. *cuneata* & *longibracteata* only) 45 (50,9) 61 μm (subsp. *cuneata*: Esterhuysen 5550; subsp. *longibracteata*: Dahlstrand 3157; subsp. *hoerleiniana*: Dinter H. 60401).
dinteri S. Moore: two-colporate, areolate, 1–2 rows areolae, length 32 (37,2) 43 μm (Edwards 4338, Smith 1339).
elegantula S. Moore: shortly two-colporate, areolate, one row areolae, length not measured (Salubeni 778).
extensa T. Anders.: three-colporate, margocolpus entire, length not measured (Leeuwenberg 6841).
flava (Vahl) Vahl: three-colporate, areolate, one row areolae, length 42 (52,3) 58 μm (Werdermann & Oberdieck 1744) (Figure 3D, E).
glabra Koenig ex Roxb.: three-colporate, margocolpus entire, length 41 (43,4) 50 μm (Smith 1700, 2734). (Figure 2D, E).
goetzei Lindau: two-porate, areolate, one row areolae on either side of pore and a single row where colpus would usually be, length not measured (Semsei 1657).
guerkeana Schinz: two-colporate, margocolpus entire, length 35 (39,3) 45 μm (Barnard 122) (Figure 4A, B, C).

- interrupta* C.B. Cl.: three-colporate, margocolpus entire, length not measured (R.B. & A.J. Faden 74/337).
- kirkiana* T. Anders.: three-colporate, areolate, one row areolae, length 47 (54,2) 61 μm (Muir 1090, Drummond 5329) (Figure 3A, C).
- minima* A. Meeuse: three-colporate, areolate, two rows areolae, length 30 (33,3) 39 μm (Immelman 145).
- montis-salinarum* A. Meeuse: three-colporate, margocolpus entire, length 37 (41,8) 45 μm (Van Wyk 5536).
- odora* (Forssk.) Vahl: two-colporate, areolate, one row areolae, length 37 (41,8) 45 μm (H. & E. Wanntorp 414).
- orchiooides* L. f. (both subspp.) two-colporate, margocolpus entire, length 30 (39,6) 49 μm (subsp. *orchiooides*: Hafstrom & Acocks H 905; subsp. *glabrata* Immelman: Muir 1100).
- petiolaris* (Nees) T. Anders. (all three subspp.): three-colporate, areolate, one row areolae, length 56 (67) 77 μm (subsp. *petiolaris*: McMurtry 1532; subsp. *incerta* (C.B. Cl.) Immelman: Meeuse 9100; subsp. *bowiei* (C.B. Cl.) Immelman: Marriott 3) (Figure 3B).
- parvibracteata* Immelman: two-colporate, areolate, 1–2 rows areolae, length 35 (37,9) 42 μm (Leistner 1402, 1657).
- platysepala* (S. Moore) P.G. Mey.: two-colporate, margocolpus entire, length 32 (37,4) 44 μm (Giess, Volk & Bleissner 5863) (Figure 5C).
- protracta* (Nees) T. Anders. (both subspp.): two-colporate, areolate, 1–2 rows areolae, length 30 (41,9) 48 μm (subsp. *protracta*: Hall 4563; subsp. *rhodesiana*: Germishuizen 974).
- stachyarrhetoides* C.B. Cl.: three-colporate, margocolpus entire, length not measured (Tinley 2624).
- thymifolia* (Nees) C.B. Cl.: two-colporate, margocolpus entire, length 53 (60,6) 68 μm (Oliver, Tölken & Venter 61).

Siphonoglossa

- leptantha* (Nees) Immelman (both subspp.): two-colporate, areolate, 2–3 rows areolae, length 58 (64,4) 77 μm (subsp. *leptantha*: Galpin 7752, 10856; subsp. *late-ovata* (C.B. Cl.) Immelman: Fourcade 3724) (Figure 5A, B, D, E, F).
- linifolia* (Lindau) C.B. Cl.: two-colporate, areolate, 2–3 rows areolae, length 74 (83,4) 90 μm (Meeuse 10093).
- nkandlaensis* Immelman: two-colporate, areolate, three rows areolae, length 71 (79,4) 88 μm (Wells 2495).

Key to pollen of *Justicia* and *Siphonoglossa* in southern Africa

- Pollen longer than 58 μm , with 2–3 rows areolae ... *Siphonoglossa*
 Pollen usually shorter than 58 μm or, if longer, then with
 one row of areolae or margocolpus entire *Justicia*

Key to pollen of sections of *Justicia* in southern Africa (Sectional delimitation follows Graham 1989)

- 1a Pollen 2-porate or 2-colporate:
 2a Pollen with entire margocolpus sect. *Justicia* p.p.
 (*J. orchiooides*, *J. cuneata*, *J. thymifolia*, *J. guerkeana*, *J. platysepala*)
 2b Pollen with areolate margocolpus:
 3a Pollen usually 2-porate (in southern African species),
 22–36 μm long .. sect. *Rostellularia* subsect. *Ansellia* p.p.
 (*J. anselliana*, *J. anagalloides*, *J. crassiradix*)
 3b Pollen 2-colporate, 34–60 μm long sect. *Harnieria*
 (*J. protracta*, *J. dinteri*, *J. parvibracteata*, *J. capensis*, *J. odora*)
 1b Pollen 3-colporate:
 4a Pollen with entire margocolpus sect. *Raphidospora*
 (*J. glabra*, *J. campylostemon*)
 sect. *Justicia* p.p.
 (*J. bolusii*)
 sect. *Betonica*
 (*J. betonica*, *J. montis-salinarum*)
 4b Pollen with areolate margocolpus:
 5a Pollen shorter than 40 μm
 sect. *Rostellularia* subsect. *Ansellia* p.p.
 (*J. minima*)
 5b Pollen longer than 40 μm sect. *Tyloglossa*
 (*J. flava*, *J. kirkiana*, *J. petiolaris*)

CONCLUSION

A number of different pollen types were found in the southern African species of *Justicia*. Either two or three colpi were found, and the margocolpus was either entire or broken up into circular areolae. Most species examined had an elongated colp, except for two southern African species of *Justicia* where the colp was lacking, one tropical African species where it was short, and another tropical African species where it was replaced by an extra row of areolae. It was possible to divide the southern African species of *Justicia* from *Siphonoglossa* on pollen characters, and also to partially divide the southern African species of *Justicia* into sections on the same basis.

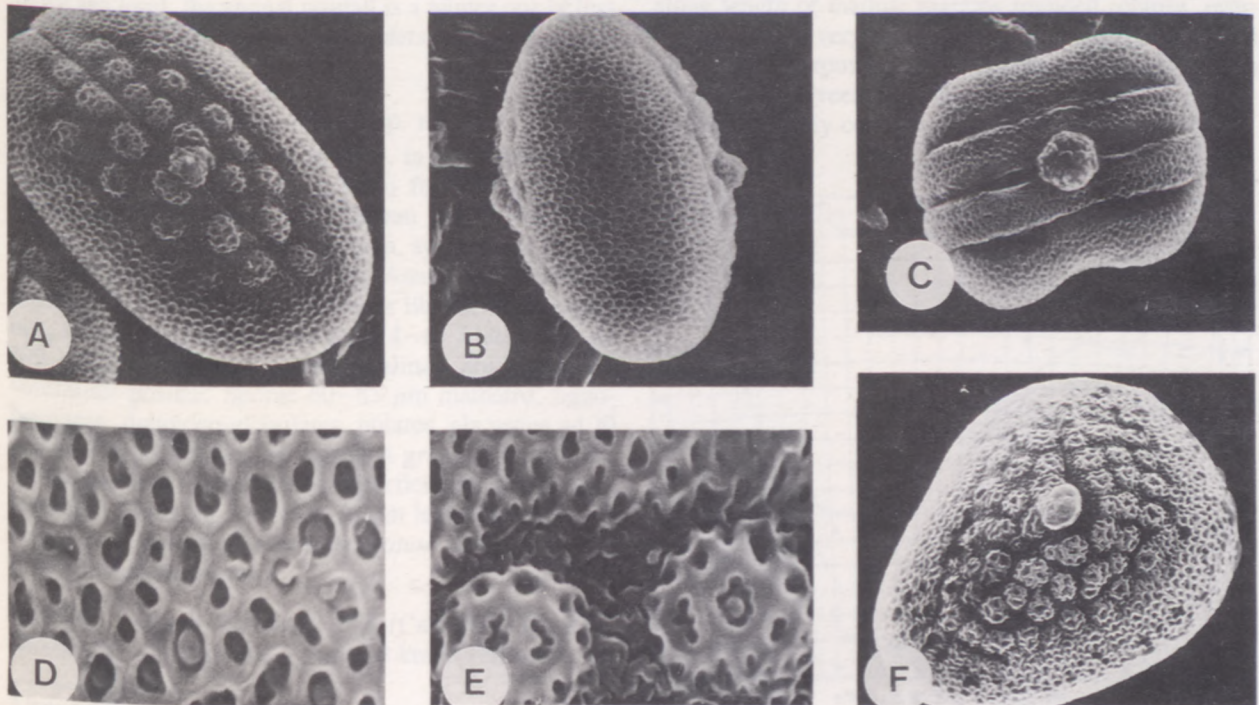


FIGURE 5.—Pollen of *Justicia* and *Siphonoglossa* species, 2-colporate, margocolpi areolate. A, B, D–F, *Siphonoglossa leptantha* subsp. *leptantha*, Galpin 7752: A, face, $\times 900$; B, side, $\times 900$; D, surface detail, $\times 5200$; E, surface detail showing areolae, $\times 5200$; F, abnormal trilete grain, $\times 900$. C, *J. platysepala*, Giess, Volk & Bleissner 5863, face, $\times 1000$.

The South African species of *Siphonoglossa* had two-colporate pollen with areolae and long colpi. On the basis of the pollen characters the genus would seem to most resemble *Justicia* sect. *Harnieria*.

ACKNOWLEDGEMENTS

This work was carried out as part requirement for a Ph.D. thesis in the Department of Botany, University of Natal, Pietermaritzburg. I would like to thank my supervisor, Dr F. Getliffe Norris, for her help and advice in the writing up of my thesis, and Mrs S. Perold of the Botanical Research Institute, Pretoria, for her patient assistance with the Scanning Electron Microscope and the photography.

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