A contribution to the floral anatomy of *Leptospermum laevigatum*

C.T. Johnson

Jniversity of Transkei, Umtata, Transkei

description of the floral parts is given. The anther is haracterized by a prominent oil gland at the tip of the conective. The structure of the seed coat of the fertile and sterile eed differs. Most of the integumental tissue is destroyed uring development and only the epidermis of the inner and uter integuments develops into the seed coat. A brief descripon of the vascularization of the flower and its floral ontogeny also given.

. Afr. J. Bot. 1983, 2: 202-208

Beskrywing van die blomdele word gegee. Die helmknop ord gekenmerk deur die prominente olieklier op die punt van ie helmbindsel. Die saadhuid van die vrugbare en steriele aadknop verskil. Slegs die epidermis van die buitenste en bineste integumente bly in die saadhuid behoue. 'n Kort eskrywing van die bearing van die blom en sy ontogenie word ok gegee.

-Afr. Tydskr. Plantk. 1983, 2: 202 – 208

eywords: Leptospermum laevigatum, flower

T. Johnson liversity of Transkei, Private Bag X 5092, ntata, Transkei

Introduction

Although the general morphology of the Myrtaceae has been investigated repeatedly and reported in many taxonomic publications, detailed studies of the internal structure and ontogeny of the flower of the genus *Leptospermum* do not exist.

The last reasonably comprehensive account of the floral morphology of *Leptospermum* was by Bentham (1866). A publication by Erdtman (1971) contains data on pollen morphology.

Material and Methods

Material of *Leptospermum laevigatum* (Soland. ex Gaertn.) F. Muell was collected from introduced but naturalized plants from various localities in the South Western Cape. Voucher specimens have been deposited in the herbarium of the University of the Western Cape.

Mature and developing flowers were fixed in F.A.A. (Johansen 1940). Material for histological examination was dehydrated in a tertiary butyl alcohol series (Johansen 1940) and embedded in paraplast with a melting point of 60 °C. Floral buds were sectioned serially at $15-24 \,\mu\text{m}$ and stained with Astra blue and Safranin. Photomicrographs were made with a 'Wild stereomicroscope and Leitz photomicroscope using 35 mm Panatomic X film.

Observations

Floral morphology

Inflorescence. Although the flowers are solitary, one occasionally finds a cyme. The flower, although described as sessile or nearly so (Bentham 1866), has a pedicel of 2-3 mm. Each flower is subtended by bracts which imbricate and bracteoles which cohere. Both bracts and bracteoles are deciduous.

Perianth. Each flower has five free sepals and five free petals which imbricate in the bud. The calyx, while in the bud stage, is pubescent, but is glabrous at a later stage. The sepals and petals are inserted on the outside of the circular hypanthium ledge.

Stamens. Numerous stamens are free and inserted in a single row on the inside of the hypanthium ledge (Figure 12f). Anthers are dorsifixed but versatile with four microsporangia which dehisce longitudinally.

A single prominent oil gland occurs at the tip of the connective (Figure 1h). In surface view, the epidermal cells overlying the gland appear as a disc between the two anther lobes. These epidermal cells are large, polygonal and к) t Nova Hedwigia Suppl. 64: 193-206.

- EHRLICH, A. 1973. Quaternary diatoms of the Hula Basin (Northern Israel). *Geol. Survey Israel, Bull.* 58: 1-39.
- GEISSLER, U., GERLOFF, J., HELMCKE, J.-G., KRIEGER, W. & REIMANN, B. 1963. In: Helmcke, J.-G. & Krieger, W. 1953-, Diatomeenschalen im elektronenmikroskopischen Bild, Part 4, plates 301-413. J. Cramer, Weinheim.
- GERLOFF, J., NATOUR, R.M. & RIVERA, P. 1978. Diatoms from Jordan. Willdenowia 8: 261-316.
- HUSTEDT, F. 1922. Bacillariales aus Innerasien. In: Southern Tibet, ed. Hedin, S., Vol. 6(3), Botany: 107-152, plates 9, 10. Lithographic Institute of the General Staff of the Swedish Army, Stockholm.
- HUSTEDT, F. 1930. Bacillariophyta (Diatomeae). In: Die Süsswasser - Flora Mitteleuropas, ed. Pascher, A., Part 10. G. Fischer, Jena, 466 pp.
- HUSTEDT, F. 1931 59. Die Kieselalgen. In: Kryptogamen Flora von Deutschland, Österreich und der Schweiz, ed. Rabenhorst, L., Vol. 7, Part 2, 845 pp. Akademische Verlagsgesellschaft Geest & Portig K.-G., Leipzig.
- HUSTEDT, F. 1937 38. Systematische und ökologische Untersuchungen über die Diatomeen-Flora von Java, Bali und Sumatra. Systematische Teil. Arch. Hydrobiol. Suppl. 15: 131 - 177, 187 - 295, 393 - 506.
- HUSTEDT, F. 1949. Diatomeen von der Sinai-Halbinsel und aus dem Libanon Gebiet. Hydrobiologia 2: 24 55.
- HUSTEDT, F. 1955. Neue und wenig bekannte Diatomeen 8. Abh. naturw. Ver. Bremen 34: 47-68.
- JACKSON, D.C. & LOWE R.L. 1978. Valve ultrastructure of species of the diatom genera *Gyrosigma* and *Pleurosigma* from the Portage River drainage system, Ohio. *Trans. Am. microsc. Soc.* 97: 569-581.
- KRAMMER, K. 1979. Zur Morphologie der Raphe bei der Gattung Cymbella. Nova Hedwigia 31: 993-1029.
- KRAMMER, K. 1980. Mikroskopie und Fotografie von Kieselalgen 1. Vergrösserung und Auflösung. Mikrokosmos 69(7): 226-233.
- KRAMMER, K. 1981a. Zur Deutung einiger Schalenstrukturen bei pennaten Diatomeen. Nova Hedwigia 35: 75-105.
- KRAMMER, K. 1981b. Morphologic investigations of the valve and girdle of the genus Cymbella Agardh. Bacillaria 4: 125-146.
- OKUNO, H. 1974. Freshwater diatoms. In: Helmcke, J.-G., Krieger, W. & Gerloff, J. 1953-, Diatomeenschalen im elektronenmikroskopischen Bild, Part 9, 1-45, plates 825-923. J. Cramer, Vaduz.
- PATRICK, R. & REIMER, C.W. 1966. The diatoms of the United States exclusive of Alaska and Hawaii, Vol. 1. Monogr. Acad. Nat. Sci. Philad. 13, 688 pp.
- PATRICK, R. & REIMER, C.W. 1975. The diatoms of the United States exclusive of Alaska and Hawaii. Vol. 2. Part 1. Monogr. Acad. Nat. Sci. Philad. 13, 213 pp.
- REICHARDT, E. 1981. Die Diatomeenflora quelliger und sumpfiger Standorte in der Umgebung von Treuchtlingen. Nova Hedwigia 34: 487-519.

- ROSS, R., COX, E.J., KARAYEVA, N.I., MANN, D.G., PAD-DOCK, T.B.B., SIMONSEN, R. & SIMS, P.A. 1979. An amended terminology for the siliceous components of the diatom cell. *Nova Hedwigia* Suppl. 64: 513-533.
- SCHMIDT, A. et al. 1874 1959. Atlas der Diatomaceenkunde. Continued by Schmidt, M., Fricke, F., Heiden, H., Müller, O. & Hustedt, F., 480 plates. O.R. Reisland, Leipzig.
- SCHOEMAN, F.R. 1969. Diatoms from the Orange Free State (South Africa) and Lesotho. No. 2. Revta Biol. 7: 35-74.
- SCHOEMAN, F.R. 1970. Diatoms from the Orange Free State (South Africa) and Lesotho. No. 3. *Botanica mar.* Suppl. 13: 49-72.
- SCHOEMAN, F.R. 1973. A systematical and ecological study of the diatom flora of Lesotho with special reference to the water quality, 355 pp. and 10 plates. V & R Printers, Pretoria.
- SCHOEMAN, F.R. 1982. The diatoms of the Jukskei Crocodile River system (Transvaal, Republic of South Africa): a preliminary check-list. Jl. S. Afr. Bot. 48: 295-310.
- SCHOEMAN, F.R. & ARCHIBALD, R.E.M. 1976. The diatom flora of southern Africa. No. 1, September 1976. CSIR Special Report WAT 50. No page numbers; series of plates with text, 43 pp. Graphic Arts Division of the CSIR, Pretoria.
- SCHOEMAN, F.R. & ARCHIBALD, R.E.M. 1977. The diatom flora of southern Africa. No. 3, September 1977. CSIR Special Report WAT 50. No page numbers; series of plates with text, 61 pp. Graphic Arts Division of the CSIR, Pretoria.
- SCHOEMAN, F.R. & ARCHIBALD, R.E.M. 1978. The diatom flora of southern Africa. No. 4, April 1978. CSIR Special Report WAT 50. No page numbers; series of plates with text, 68 pp. Graphic Arts Division of the CSIR, Pretoria.
- SCHOEMAN, F.R. & ARCHIBALD, R.E.M. 1979. The diatom flora of southern Africa. No. 5, September 1979. CSIR Special Report WAT 50. No page numbers; series of plates with text, 65 pp. Graphic Arts Division of the CSIR, Pretoria.
- SCHOEMAN, F.R. & ARCHIBALD, R.E.M. 1980. The diatom flora of southern Africa. No. 6, May 1980. CSIR Special Report WAT 50. No page numbers; series of plates with text, 111 pp. Graphic Arts Division of the CSIR, Pretoria.
- SCHOEMAN, F.R. & ASHTON, P.J. 1982a. The diatom flora of the Pretoria Salt Pan, Transvaal, Republic of South Africa. Bacillaria 5: 63-99.
- SCHOEMAN, F.R. & ASHTON, P.J. 1982b. The diatom flora in the vicinity of the Pretoria Salt Pan, Transvaal, Republic of South Africa I. Nova Hedwigia Suppl. 73: 21-54.
- VAN HEURCK, H. 1880-83. Synopsis des Diatomées de Belgique. Atlas: 132 plates. J. Ducaju & Co., Anvers.
- VAN HEURCK, H. 1884-87. Types du Synopsis des Diatomées de Belgique, Série I-XXII, 123 pp., 550 slides. Anvers.
- VAN HEURCK, H. 1885. Synopsis des Diatomées de Belgique. Text: 235 pp. and 3 plates. J. Ducaju & Co., Anvers.
- VANLANDINGHAM, S.L. 1969. Catalogue of the fossil and recent genera and species of diatoms and their synonyms, Part 3: *Coscinophaena* through *Fibula*, p. 1087-1756. J. Cramer, Lehre.



gures 70-77 Figures 70-76. Gyrosigma rautenbachiae Cholnoky, sample B2, SEM internal views. 70. centre of valve. Note the axial rib within the ial groove (× 4000). 71. central nodule of specimen in 70 enlarged and turned. Note the two, short, longitudinal costa (× 6000). 72. part of a valve owing the axial rib (AR), helictoglossa (H) and tooth-like projections (P) (× 4000). 73. pole of a specimen in 72 turned and tilted to illustrate the ped helictoglossa (× 6000). 74. pole of another valve seen from a different angle (× 6000). 75. pole of a valve showing the internal raphe fissure F emerging onto the crest of the axial rib and terminating on the helictoglossa (× 6000). 76. cross-section of a valve showing the double-walled ucture in which the striae are composed of loculate areolae. Note the tooth-like projections (P) (× 6000).

gure 77. Gyrosigma rautenbachiae Cholnoky, Pienaar's River (PR 11), Transvaal. Cholnoky type material. SEM, external view of cross-section through lve. The slight lateral displacement of the valve wall along the raphe shows that the raphe has the form of a deep tongue and groove structure (\times 6000).

icroscopes has revealed that our specimens of *Cymbella icrocephala* Grunow should be divided into two groups. his has been discussed in the light of the available literature nd comparisons have been made with examples of this becies from the van Heurck Type Slide No. 211. Unforinately we were unable to obtain unmounted type material f *C. microcephala* for electron microscopy. Nevertheless, ir observations have focussed attention on this problem.

We have also been able to compare our specimens of *yrosigma rautenbachiae* Cholnoky with Cholnoky's type aterial. This has enabled us to furnish light microscope lotographs and electron micrographs for this species which as previously only been illustrated in the literature by a w line drawings.

A number of other taxa are also listed for the record but o not require further comment as they are well known.

cknowledgements

his paper is published with the approval of the Director the National Institute for Water Research. Our sincere anks are due to Dr R.E.M. Archibald for his valuable iticism of this manuscript, and Mrs V.H. Meaton for the final photographic prints. We are also indebted to Dr O. Almborn of the Botanical Museum in Lund, Sweden, and Dr. U. Passauer of the Naturhistorisches Museum in Vienna for their willingness to assist us with the loan of diatom slides and exsiccati.

References

- AGARDH, C.A. 1830. Conspectus criticus Diatomacearum. Part 1. Litteris Berlingianis, Lundae, 16 pp.
- ANONYMOUS 1975. Proposals for a standardization of diatom terminology and diagnoses. *Nova Hedwigia* Suppl. 53: 323 – 354.
- CHOLNOKY, B.J. 1955. Diatomeen aus salzhaltigen Binnengewässern der westlichen Kaap — Provinz in Südafrika. Ber. dt. bot. Ges. 68: 11-23.
- CHOLNOKY, B.J. 1957. Beiträge zur Kenntnis der südafrikanischen Diatomeenflora. Port. Acta biol. (B) 6: 53-93.
- CHOLNOKY, B.J. 1959. Neue und seltene Diatomeen aus Afrika. IV. Diatomeen aus der Kaap — Provinz. Öst. bot. Z. 106: 1-69.
- COX, E.J. 1977a. Variation in valve structure between species of the diatom genus Cymbella C.A. Ag. Nova Hedwigia 28: 427-449.
- COX, E.J. 1977b. Raphe structure in naviculoid diatoms as revealed by the scanning electron microscope. Nova Hedwigia Suppl. 54: 261-274.
- COX, E.J. 1979. Symmetry and valve structure in naviculoid diatoms.

cf. Okuno 1974, plate 864; Cox 1979, plate 2, figure 10) and a slit like aperture to the exterior (Figure 77). Figure 77 provides a particularly good illustration of the raphe structure. A slight lateral displacement of the valve wall along the raphe shows that it has the form of a deep tongue and groove structure. This can be compared to similar structures in *Stauroneis phoenicenteron* and *Pinnularia microstauron* (cf. Krammer 1981a, plate 3, figures 18, 19 and plate 5, figure 42 respectively). A small number of G. rautenbachiae valves were recorded from sample B2.

Conclusions

With the aid of type material we have been able to establish the true identity of *Cymbella cymbiformis* Agardh, a species whose taxonomy has previously been much confused.

Critical examination under both light and electron



Figures 61 - 69 Figures 61 - 63. Gyrosigma rautenbachiae Cholnoky, sample B2, LM. 62 & 63. same specimen but different illumination in 63 to reveal the raphe branches (\times 500).

Figure 64. Gyrosigma rautenbachiae Cholnoky, Riet Stream near Nelspruit (Eastern Transvaal), sample ET 30 (NIWR Slide No. 206/4105), L.M. Focused to show the straight, internal, proximal raphe ends (× 1100).

Figures 65 & 66. Gyrosigma rautenbachiae Cholnoky, Crocodile River above Hartbeespoort Dam, Transvaal (NIWR Slide No. 123/2458), LM. 66. specimen in 65 enlarged to show the straight, internal, proximal raphe ends (65. × 500; 66. × 1750).

Figure 67. Gyrosigma rautenbachiae Cholnoky, Hennops River, Transvaal, SEM. External view of the centre of an etched valve. Note the curved proximal raphe ends (× 5000).

Figures 68 & 69. Gyrosigma rautenbachiae Cholnoky, sample B2, SEM. Internal views showing the central nodule (CN), axial groove (AG), axial rib (AR), internal raphe fissure (IRF), longitudinal costa (C) and tooth-like projections (P) (× 4000).

Figures 61, 63 & 65. oblique - bright field illumination. Figures 62, 64 & 66. bright field illumination.

S.-Afr. Tydskr. Plantk., 1983, 2(3)

100

·~3

ngated pores thereby resembling more closely our group xamples. A further interesting feature in the group 2 ximens is the lip-like extension of the valve mantle at the les (Figures 57, 58) which may be a diagnostic characteic of this group.

ł

Finally, internal views of valves from both groups gures 54, 55, 57, 58) displayed a slightly raised central fule (not much expanded laterally) and a raised axial ge. Small helictoglossae were observed in group 2 valves gures 57, 58). It is evident from this study that further I studies on authenticated material of *C. microcephala* necessary to establish the true identity of this taxon. Untunately we have been unable to obtain unmounted type terial for EM examination. Our studies have, neverthei, shown the taxonomy of this species to be more comx than is generally known.

The value dimensions for the two groups of C. rocephala as distinguished in this study are as follows:

- bup 1: Length $14,0-22,5 \ \mu\text{m}$, breadth $3,3-4,4 \ \mu\text{m}$. Transapical striae $24-26 \text{ in } 10 \ \mu\text{m}$ dorsally (off centre), $22-26 \text{ in } 10 \ \mu\text{m}$ ventrally and $27-30 \ \text{in } 10 \ \mu\text{m}$ at the poles (dorsally and ventrally).
- Sup 2: Length $13,7-19,0 \ \mu\text{m}$, breadth $3,4-4,4 \ \mu\text{m}$. Transapical striae 26-32 in $10 \ \mu\text{m}$ (usually a little less dense on the ventral side) and 28-34 in $10 \ \mu\text{m}$ at the poles (dorsally and ventrally).

cription of *Cymbella microcephala* Grunow based on Heurck's Type Slide No. 211:-

valves vary in shape from linear-lanceolate to almost pletely linear forms, $12,5 - 16,5 \mu m$ long and 3,4 - 4,0broad. The poles are protracted with rostrate, subitate or capitate apices. Some valves have a convex dornargin and an almost straight ventral margin. The raphe nches appear robust and arcuate with the convex side cted towards the ventral valve margin. The central pores not conspicuous and the hooked terminal fissures are ed near the dorsal margin of the pole and curve down ards the ventral margin. The axial area is narrow, owing the curvature of the raphe branches and shows ctically no expansion at the centre. A true central area erefore absent. The transapical striae vary from radial ughout to slightly radial or parallel at the poles. isured along the raphe the striae number (20) 24-28 $0 \,\mu m$ near the centre on the dorsal side, 20 - 26 in $10 \,\mu m$ g the ventral side and 24 - 30 in 10 μ m at the poles (veny and dorsally).

osigma rautenbachiae Cholnoky (Figures 61 – 77) lnoky 1957, 65, figure 61.

gth 155,0 – 175,0 μ m, breadth 25,0 – 26,0 μ m. Transal striae 13 – 14 in 10 μ m, longitudinal striae about 15 0 μ m.

his taxon appears to be more common than originally ight and has been recorded by the senior author from ral localities in the Transvaal (Schoeman 1982, 301, as p. 1 affin. *rautenbachiae*; various unpublished data). specimens from the vicinity of the Pretoria Salt Pan carefully compared with Cholnoky's Type material in the Pienaar's River, Transvaal (Cholnoky 1957, 65), found to be identical in all respects. Examination of other authenticated Cholnoky material for this taxon revealed that in certain cases a species having a strong resemblance to *G. acuminatum* (Kützing) Rabenhorst had been misidentified as *G. rautenbachiae* Cholnoky.

In general appearance and shape, as seen under the light microscope, G. rautenbachiae Cholnoky (Figures 61 – 63, 65) resembles G. acuminatum (Kützing) Rabenhorst (Hustedt 1930, 222, figure 329; Patrick & Reimer 1966, 314, plate 23, figures 1–3) and G. attenuatum (Kützing) Rabenhorst (Hustedt 1930, 224, figure 330; Patrick & Reimer 1966, 319, plate 24, figure 1). It is distinguished from G. acuminatum by its broader valves and by possessing fewer transapical and longitudinal striae in 10 μ m. Gyrosigma rautenbachiae differs from G. attenuatum in the number of longitudinal striae in 10 μ m whereas G. rautenbachiae usually has 14–15 in 10 μ m.

In LM the internal and external proximal ends of the raphe fissures in G. rautenbachiae can be resolved by focusing in two planes. Internally, the proximal ends of the raphe fissure are straight (Figures 64, 66), while externally, the proximal ends of the raphe fissure are hooks curving in opposite directions. These external hooked proximal raphe endings are clearly visible under SEM (Figure 67; cf. G. acuminatum sensu Jackson & Lowe 1978, figure 3) and extend across two rows of the longitudinal striae on either side of the central nodule. They differ from those of G. attenuatum sensu Cox (1977b, plate 4, figure 31; 1979, plate 1, figure 1) which appear sinuate in the curved portion and do not cut across the striae. Internally the raphe fissures extend only a short distance onto the central nodule and end abruptly in slightly expanded pores (Figure 68). Figure 67 shows the arrangement of the pores in intersecting traverse and longitudinal striae. The raphe runs in an axial rib which is flush with the valve surface on the outside (Figure 67) but internally is a ridge lying within a groove (Figures 69, 71, 74, 76). The external (Figure 67) and internal (Figures 69, 76) raphe fissures run along the side of the axial rib except at their internal proximal and distal ends where they emerge on the crest of the rib (Figures 68, 75). In the central parts of the valve the groove containing the axial rib appears to be rather deep (Figures 70, 71) but towards the poles it becomes shallower so that the rib often projects above the inner valve surface (Figures 73, 74, 75). Internally at the centre of the valve the axial rib is expanded into a spindle-shaped central nodule (Figures 68, 71). On either side of this central nodule there is a short longitudinal costa (both of equal length) which does not extend further than this region (Figures 68-71; cf. G. attenuatum sensu Cox 1979, plate 1, figure 8). It is interesting to note that Cox illustrates G. attenuatum with both uneven (Cox 1977b, plate 4, figure 29) and even (Cox 1979, plate 1, figure 8) length axial costae. Further towards the poles, along the edge of the axial groove on the side opposite that facing the opening of the raphe fissure in the axial rib, there is a row of tooth-like projections (Figures 72, 74, 76; cf. Cox 1979, plate 1, figure 8). This is clearly demonstrated in Figure 76. The well developed terminal nodule takes the form of a lipped structure or helictoglossa in which the distal ends of the raphe terminate (Figures 73 - 75). In Figures 76 and 77 the valve is also shown as a double walled structure having a thin outer wall and a slightly thicker inner wall. The striae are comprised of loculate areolae with a more or less rectangular occluded inner opening (Figure 76:

appear to lie in a groove with a crenulate margin. This structure is clarified when valves are viewed under the SEM. The slit-like pores are then seen to represent the outer pore openings on the surface of the valve, while the internal pore openings are round (cf. Ehrlich 1973, plate 7, figure 6) thereby creating the crenulate image observed under TEM. The round internal pore openings lie in narrow grooves which are separated by transapical costae (Figures 54-56).

Under TEM the striae of the group 2 specimens (Figures 52, 53) consist of round to oblong-shaped pores $(40-44 \text{ in } 10 \ \mu\text{m})$ which lie within very narrow grooves that possess

straight (non-crenulate) sides (Figure 53; cf. Figures 59, 60 as examples from Lesotho previously identified as *C. microcephala*). When viewed (under SEM) from the inside of the valve (Figures 57, 58) these straight-sided grooves (group 2 specimens) are found to be narrower than those of the group 1 valves. The structure of the outer valve surface (group 2) as seen under SEM is basically similar to that illustrated by Ehrlich (1973, plate 7, figure 5) and Gerloff *et al.* (1978, plate 11, figure 3). The illustration of Krammer (1981b, plate 1, figure 7) of the outer surface of a *C. microcephala* valve depicts both round and transapically



Figures 52 – **60** Figures 52 & 53. *Cymbella microcephala* Grunow (group 2), TEM. 53. upper pole of specimen in 52 enlarged (52. × 3500; 53. × 11400) Figures 54 & 55. *Cymbella microcephala* Grunow (group 1), SEM, internal views of the same specimen but valve tilted in 55 (× 10000). Figure 56. *Cymbella microcephala* Grunow, SEM micrograph of internal view by kind permission of Dr Kurt Krammer, Meerbusch, West Germany (× 15000) Figures 57 – 60. *Cymbella microcephala* Grunow (group 2), 57 & 58. SEM, internal views (× 6000). 59 & 60. TEM, specimen from Lesotho (59. × 8400). 60a & b. part of specimen in 59 enlarged but taken at different exposure times to illustrate the striae structure in 60a and raphe detail in 60b (× 17300)

٢

b

ent of inner and outer raphe fissures from the perpencular plane. Grunow's drawings in van Heurck (1880-83, ate 8) illustrate a slight dorsal curvature of the raphe anch in his Figures 36 and 37 and a ventral one in his gure 39! According to Krammer's classification (1979, 05, plate 3, figure 22) our specimens belong to the group *Cymbellae* possessing lateral raphe systems. Our TEM crographs also clearly illustrate the narrow axial area ich follows the curvature of the raphe branches. A cenul area is absent, though occasionally there may be a slight dening around the central nodule. The terminal areas are tall.

Cox (1977a, 431, plate 3, figures 13 - 15) recorded three types of striae pores within the genus *Cymbella*. The pore structure of our specimens does not fit any of her types. Under TEM we observed two distinct transapical striae types thereby lending support to the division of our specimens into two *C. microcephala* groups. The striae of specimens in our group 1 (Figures 47 - 51) contained 36 - 41 puncta in 10 μ m. These puncta are linear or slightly dumbbell in shape and are aligned with their long axes parallel to the striae. Near the valve apices a few pores occasionally assume a trifid shape. Under TEM we also observed that in these specimens (Figure 51) the transapically aligned slit-like pores



Ires 29–51 Figures 29–34. *Cymbella microcephala* Grunow (group 1), LM. 29 & 30. same specimen (× 1500). **Ires 35–38.** *Cymbella microcephala* Grunow (group 2), LM (× 1500).

tres 39-46. Cymbella microcephala Grunow, Bruxelles, Belgium. Van Heurck Type Slide No. 211 = NIWR Slide No. 11/211. 39-41. camera la drawings (*delin*. R.E.M. Archibald). 42-46. LM. 45 & 46. same specimen with 45 illustrating the transapical striae and 46 the valve outline (× 1500). tres 47 & 48. Cymbella microcephala Grunow (group 1). TEM (47. × 3200; 48. × 6400). tres 49-51. Cymbella microcephala Grunow (group 1), TEM. 51. part of specimen in 50 enlarged to show the structure of the transapical striae

& 50. × 5400; 51. × 18000).

res 29, 36, 38 & 42-46. phase contrast illumination; Figures 30-35 & 37. oblique - bright field illumination.



Figures 19-28 Figures 19-23. Cymbella cymbiformis Agardh, SEM. Examples from sample B2. 19 & 20. external views showing a valve centre (19) with two stigmata, and a pole (20) with distal raphe end and mucilage pore area (MP) (\times 6000). 21-23. internal views. 21. whole valve of a small specimen (\times 1000). 22. valve centre with central nodule and inner openings of the stigmata (\times 3250). 23. centre part of specimen in 22 tilted and enlarged to show the stigmata (S) (\times 4680).

Figures 24 – 27. Cymbella cymbiformis Agardh, sample B2, SEM internal views. 24. centre with raised axial rib, thickened central nodule and two stigmata (\times 3250). 25 – 27. poles of different specimens. Note the helictoglossa (H), the dorsal thickened extension (E) of the terminal nodule and the terminal mucilage pore area (25. \times 6000; 26 – 27. \times 4680).

Figure 28. Cymbella cymbiformis Agardh, sample B2, TEM. Enlargement of valve apex to show the striae structure and the striate nature of the terminal mucilage pore area (MP) (× 4680).

of the structure can be seen, though in valve outline they do resemble this taxon. Grunow's first reference to C. microcephala is to be found in van Heurck's Atlas (1880-83), plate 8, figures 36-39) of the diatoms of Belgium, and van Heurck (1885, 63) cited his Type Slide No. 211 as containing this species. Since Grunow (in van Heurck 1884-1887, series 9) substantiated the identity of C. microcephala on van Heuck's Type Slide No. 211 we may thus assume it is either the syntype slide or properly authenticated material for this taxon. We examined this slide (obtained from the Grunow diatom collection in Vienna) and found that the specimens (Figures 39-46; cf. Patrick & Reimer 1975, 33, plate 4, figures 12, 13) resemble many of our group 2 valves. A description of the valves observed on this slide (No. 211) is given at the end of the discussion on C. microcephala. According to van Heurck (1885, 63) the raphe is weakly arcuate, but specimens we examined on the van Heurck Type Slide No. 211 usually had raphe branches slightly more arcuate than those illustrated by Grunow in van Heurck (1880-83, plate 8, figures 36-39). Hustedt's illustrations (1930, 351, figure 637; 1949, plate 1, figures 34-38), on the other hand, tend to depict almost straight raphe branches which come closer to our group 1 specimens.

It was only after completing an EM study on the Pretoria Salt Pan specimens (with emphasis on the transapical striae) that we found better evidence for our suspected separation of this taxon into 2 groups, as mentioned earlier. Under TEM (both groups) the raphe in valvar view (Figures 47-50, 52) is more or less centrally placed. The terminal fissures (Figures 48, 49) are directed ventrally and the small central pores sometimes show a slight dorsal deflection (Figures 47, 50, 52). The central section of each raphe branch (Figures 47-50, 52; cf. Geissler *et al.* in Helmcke & Krieger 1963, 33, plate 391, figure lower left) is ventrally curved and appears band-shaped owing to slight displace-

1.



igures 10 - 18 Figures 10 & 11. *Cymbella cymbiformis* Agardh, LM. Examples from our sample B2 (× 1000; bright field illumination). igures 12 - 18. *Cymbella cymbiformis* Agardh, SEM. Agardh (Stangby No. 4612) type material. 12 & 13. external views. 12. centre of a specimen showig stigmata (S) (× 2880). 13. pole of a valve with outer raphe fissure and mucilage pore area (MP) (× 4680). 14 - 18. internal views. 14. enlargement > show that the striae puncta open into transapically aligned grooves (× 6000). 15. internal openings of three stigmata (S) (× 4680). 16 & 17. same ole of a specimen but turned and tilted in 17. Note the helictoglossa (H) and mucilage pore area (MP) (× 6000). 18. pole of broken valve with inner aphe fissure terminating in the helictoglossa (H) (× 6000).

Lymbella cymbiformis was recorded from two samples, B1 nd B2.

symbella microcephala Grunow (Figures 29-60).

he initial examination of the slides showed this species to e the dominant taxon in samples B1 and B9. Further study nder the light microscope revealed that these specimens ould be divided into two groups on the grounds of some inor differences, namely, *C. microcephala* group 1 Figures 29-34) and *C. microcephala* group 2 (Figures 5-38). Group 1 specimens were recorded from sample B9, hile a few examples were also present in sample B1. Group specimens were present only in sample B1, where they presented the dominant diatom taxon.

Valves of group 1 have almost straight or slightly curved aphe branches whereas those in group 2 have clearly arlate and slightly thicker raphe branches. The number of orsal transapical striae (off centre) is usually between 24 ad 26 in 10 μ m for group 1 and slightly denser for group (26-32 in 10 μ m). However, the occasional intermediate becimen was inevitably present, making it extremely difficult to distinguish between the two groups. The transapical striae of specimens in both groups may be radial throughout or can become parallel or even convergent at the poles.

Examination of the available literature revealed that very little of the variation displayed by *C. microcephala* has been illustrated. Hustedt (1949, plate 1, figures 34-38) and Schoeman (1970, 52, plate 1, figures 15-22) figure some valve variation and Schoeman (1970), based on his light microscopy studies of Lesotho material, has proposed the sinking of *C. ruttneri* Hustedt (in Schmidt *et al.* 1874–1959, plate 373, figures 39-44; Hustedt 1937–38, 421, plate 25, figures 4-9; cf. Schoeman 1969, 43, figures 15-17) into the *C. microcephala* species. Electron microscopical studies of *C. ruttneri*-type material may prove otherwise.

According to VanLandingham's 'Catalogue of the fossil and recent genera and species of diatoms and their synonyms' (1969, 1212 – 1213), the earliest reference to this taxon comes under the name *Cymbella minuscula* Grunow and comprises a series of figures in Schmidt *et al.* (1874 – 1959 (1875), plate 9, figures 58 - 61). Our photographic copy of these illustrations is poor and little



Figures 1-9 Figures 1-3. Achnanthes engelbrechtii Cholnoky. 1. LM, raphe valve (\times 2000; phase contrast illumination). 2. TEM, raphe valve (\times 5900). 3. TEM, rapheless valve (\times 5900). 2 & 3. specimens from Lake Chrissie, Transvaal.

Figures 4 & 5. Amphora veneta Kützing, SEM, external views of the same valve showing the centre in 4 and a pole in 5 (× 6000).

Figures 6-9. Cymbella cymbiformis Agardh, NIWR Slide No. 422/8439 prepared from Agardh type material (Stangby No. 4612) kindly provided by Dr. O. Almborn of the Lund Botanical Museum, Sweden. LM (\times 1000; 6-8. bright field illumination; 9. oblique-bright field illumination).

12, 19). The distal ends are deflected dorsally (Figures 13, 20). The central nodule is very thickened internally (Figures 15, 24) thereby obscuring the proximal raphe endings (cf. Cox 1977a, 432). The distal ends of the axial rib are also thickened and are developed on the dorsal side into a two-lipped structure, the helictoglossa (Figures 17, 25-27) in which the inner raphe fissure terminates (Figure 18). The distal ends of the inner raphe fissures appear to run ventrally along the base of the raised axial rib before entering the helictoglossae. From the thickened terminal nodule there is a fairly broad extension towards the dorsal margin (Figures 17, 26, 27) separating the striae from the mucilage pore area (apical pore field). Cox (1977a) has suggested that

the development of a hooded raphe ending seems to be associated with the presence of a mucilage pore area at the valve apex. Externally these mucilage pores appear as rows of small closely arranged circular openings (Figure 20; cf. Cox 1977a, plate 5, figure 30) which are separated internally by narrow ribs (Figures 17, 27). On the ventral side of the central area two or occasionally three stigmata are present. Furthermore, specimens with a single stigma were observed in Agardh's-type material. Externally they appear as rather conspicuous, simple, circular openings (Figures 12, 19; cf. Krammer 1981a, plate 3, figure 20). Internally they are slit-like and are partly occluded by outgrowths of silica (Figures 15, 22-24; cf. Cox 1977a, 434).

٩.,

camera lucida with adjustable magnification changer. The original drawings were made at a magnification of $4000 \times$ and then suitably reduced for reproduction.

Additional material (slides and exsiccati) for comparative purposes was obtained from the University of Lund Botanical Museum in Sweden (for *Cymbella cymbiformis* Agardh), the Naturhistorisches Museum in Vienna (for *Cymbella microcephala* Grunow) and the NIWR diatom collection in Pretoria (for *Gyrosigma rautenbachiae* Cholnoky). Terminology used is that suggested by the Working Party on Diatom Terminology (Anon. 1975; Ross *et al.* 1979).

Observations and Discussion

A further number of diatom taxa identified from samples collected in the vicinity of the Pretoria Salt Pan are listed below in alphabetical order. Where necessary, their dimensions and some comments on their structure are also given.

Achnanthes engelbrechtii Cholnoky (Figures 1-3)

Cholnoky 1955, 16, figures 1-8.

Cholnoky 1959, 7, figures 16-33.

Length $8,0-11,0 \ \mu m$, breadth $3,5-4,2 \ \mu m$. Transapical triae (raphe and rapheless valves) 16-18 in $10 \ \mu m$ (a little lenser at the apices).

This species shows some similarity to A. pinnata Hustedt 1922, 123, plate 9, figures 15-18; 1937-38, 201, plate 13, igures 54-57) and a detailed study of Hustedt's type naterial is required to make a proper comparison. It is also netresting to note that under TEM the striae structure is eminiscent of that of A. lanceolata Brébisson ex Kützing. 1. engelbrechtii occurred only rarely in sample B9.

Chnanthes exigua Grunow

choeman & Archibald 1977, no. 3.

ichoeman & Ashton 1982a, 84, figures 1-8, 75-79, 05-110.

Length $8,0-12,0 \ \mu\text{m}$, breadth $4,3-5,0 \ \mu\text{m}$. Transapical triae: raphe valve 26-28 in $10 \ \mu\text{m}$ near the centre (slightly lenser at the poles); rapheless valve 25-27 in $10 \ \mu\text{m}$ near he centre (denser at the poles).

1. exigua was observed in samples B2 and B4.

Ichnanthes lanceolata Brébisson ex Kützing

Iustedt 1931 - 59, 408, figure 863.

'atrick & Reimer 1966, 269, plate 18, figures 1-10. A few valves were recorded from sample B2.

mphora ovalis (Kützing) Kützing var. *affinis* (Kützıg) van Heurck *ex* De Toni.

'atrick & Reimer 1975, 69, plate 13, figures 3,4. single specimen was observed in sample B2.

mphora veneta Kützing (Figures 4, 5)

choeman & Archibald 1978, no. 4.

choeman & Archibald 1979, no. 5.

length $18,0-27,5 \ \mu\text{m}$, breadth $3,2-4,0 \ \mu\text{m}$. Transapical triae on the dorsal side, punctate, 26-28 in $10 \ \mu\text{m}$, more videly spaced at the centre (about 22 in $10 \ \mu\text{m}$) and slightly enser at the poles ($28-30 \ \text{in} 10 \ \mu\text{m}$); striae along the venal side $28-30 \ \text{in} 10 \ \mu\text{m}$. A critical study of this taxon has een presented by Schoeman and Archibald (1978; 1979). Inder SEM the fairly high dorsal mantle is partially seen a Figure 4. Also clearly visible is the outer raphe fissure. dorsally to form a poorly developed conopeum (previously referred to as a flap by Schoeman & Archibald 1978; 1979). Externally the terminal fissure of the raphe is strongly recurved towards the dorsal side (Figure 5) ending in the last punctum of the most distal dorsal stria.

A. veneta was recorded from samples B1 and B5.

Cyclotella atomus Hustedt

Schoeman & Archibald 1976, no. 1. Diameter 4,0-5,0 μ m; marginal costae 13-15 in 10 μ m. This species was observed in sample B9.

Cyclotella meneghiniana Kützing

Schoeman & Archibald 1980, no. 6. A few valves were observed in sample B5.

Cymbella cymbiformis Agardh (Figures 6-28)

Length 70,0-89,0 μ m, breadth about 16,0 μ m (one specimen 12,5 μ m). Transapical striae (dorsal and ventral) about 8 in 10 μ m near the centre but denser at the poles, 12-16 in 10 μ m. The striae are punctate with 14-20 puncta in 10 μ m (20-22 in 10 μ m according to Krammer 1980, 232, figures 15,16) but denser at the poles. In our specimens two conspicuous stigmata are located on the ventral margin of the central area (Figure 10), while occasionally a third smaller stigma is also visible.

There appears to be a great deal of confusion concerning the taxonomy of this taxon and its relationship with other closely related taxa, namely, C. cistula (Hemprich) Kirchner and C. affinis Kützing (cf. Schoeman 1973, 55; Patrick & Reimer 1975, 54 - 55; Reichardt 1981, 502 - 503). This is partly due to Hustedt's (1955, 50-51) investigation and interpretation of this taxon in which he concluded that C. cymbiformis was characterized by the lack of an isolated stigma in the central area. It is evident that Hustedt (1955) did not examine any Agardh material and based his observations mainly on the Kützing material of Cocconema cymbiforme Kützing. We were fortunate to obtain some of the Agardh (1830, 10) type material from Stangby (No. 4612) for EM study from the University of Lund Botanical Museum (Sweden), together with a lectotype slide prepared from this material by Reimer (vide Patrick & Reimer op. cit.). Some illustrations of the Agardh-type specimens (Figures 6-9, 12-18) are presented here for the purpose of direct comparison with our valves (Figures 10, 11, 19-27), which were found to be identical.

Under TEM the pores of the transapical striae appear linear and are aligned with their long axes perpendicular to the striae (Figure 28). In lightly cleaned material these slitlike pores are very narrow, but harsh cleaning results in etching so that the pores appear wider and more oblong in shape. SEM examination showed that these slits represent the outer openings of the striae pores (Figures 12, 13, 19). The internal openings of these pores appear to be larger (round-oblong) and open into transapically aligned grooves (Figures 14, 22, 23).

According to Krammer's classification (1979, 1005, plate 3, figure 22, plate 4, figures 40, 41) the raphe system is of the reverse-lateral type. It lies in an axial rib which is rather flat externally (Figure 12), but appears as a raised ridge internally (Figures 15, 18, 24, 27). Viewed under SEM the external raphe fissure becomes sinuate near the valve centre with a ventrally directed arch terminating in a fairly conspicuous central pore which also bends ventrally (Figures

The diatom flora in the vicinity of the Pretoria Salt Pan, Transvaal, Republic of South Africa. Part II

F.R. Schoeman and P.J. Ashton

National Institute for Water Research, Council for Scientific and Industrial Research, Pretoria

This is the second in a series of three contributions covering the diatom flora recorded in the vicinity of the Pretoria Salt Pan. Of the ten species described in this publication, three viz. *Cymbella cymbiformis* Agardh, *C. microcephala* Grunow and *Gyrosigma rautenbachiae* Cholnoky, are discussed in detail with notes on their taxonomy and valve structure, the latter based on both light and electron microscopy. In an attempt to verify the correct identity of these taxa, certain authenticated materials were also examined.

S. Afr. J. Bot. 1983, 2: 191 - 201

Hierdie is die tweede in 'n reeks van drie publikasies oor die diatoomflora wat in die omgewing van die Pretoria Soutpan gevind is. Drie van die tien spesies wat in hierdie publikasie behandel word, naamlik *Cymbella cymbiformis* Agardh, *C. microcephala* Grunow en *Gyrosigma rautenbachiae* Cholnoky, word in besonderhede bespreek, met notas oor hul taksonomie en skaalstruktuur. Laasgenoemde is gebaseer op beide lig- en elektronmikroskopie. Om die ware identiteit van hierdie taksons te probeer vasstel, is sekere egverklaarde voorbeelde ook ondersoek.

S.-Afr. Tydskr. Plantk. 1983, 2: 191 - 201

Keywords: Diatomaceae, electron microscopy, morphology

F.R. Schoeman and P.J. Ashton

National Institute for Water Research, Council for Scientific and Industrial Research, P.O. Box 395, Pretoria 0001, Republic of South Africa

ŧ.

Introduction

The hypersaline Pretoria Salt Pan with its saline artesian spring is the only example of a maar lake in southern Africa. In a recent publication, the diatom flora of this floristically unknown system was discussed (Schoeman & Ashton 1982a). The water sources in the farmlands (the Soutpan agricultural experimental farm) surrounding the salt pan and its volcanic crater consist of a perennial stream and several boreholes whose hydrochemistry is quite different to that of the spring feeding the Pretoria Salt Pan. Since the diatom flora of the surface waters in this area was virtually unknown, the diatoms recorded from these different water sources were studied by both light and electron microscopy. The first results dealing with several taxa recorded from this area have already been published (Schoeman & Ashton 1982b).

The present paper lists additional taxa from these localities and provides dimensions, comments and comparisons with authenticated materials where these are deemed necessary. A final paper dealing with the remaining taxa will be submitted in the near future. For details on hydrochemical conditions pertaining to these sampling points, readers are referred to Schoeman & Ashton (1982b).

Study area and sampling points

The Soutpan agricultural experimental farm is located at 25°24'S and 28°05'E in the Transvaal Province, South Africa (Schoeman & Ashton 1982b) and surrounds the volcanic crater with its saline lake, the Pretoria Salt Pan. Algal samples were collected from a stream and various open water reservoirs and cattle drinking troughs associated with several boreholes on the farm (full details of these sampling points are given in Schoeman & Ashton 1982b).

These waters had relatively low concentrations of total dissolved solids (TDS) $(190-500 \text{ mg } \text{l}^{-1})$ and total alkalinity $(1,5-6,0 \text{ mequiv } \text{l}^{-1})$ and their pH values ranged from 7,0 to 7,6. In contrast, the corresponding values for the artesian springs were 3 380 mg l⁻¹, 12 m-equiv l⁻¹ and 8,8 while those for the surface waters of the Pretoria Salt Pan were 78 830 mg l⁻¹, 390 mequiv l⁻¹ and 10,4 respectively.

Materials and Methods

Methods for the preparation and examination of the diatom samples (light and electron microscopy) have also previously been presented (Schoeman & Ashton 1982b). The line drawings (Figures 14 - 16) were made with the aid of a Zeiss