Revision of some spathidiid genera (Alveolata, Ciliophora, Spathidiida)

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Revision of some spathidiid genera (Alveolata, Ciliophora, Spathidiida)

Edited by

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Cover: *Epispathidium papilliferum* (front; see Fig. 6.11h–j in Chapter 6); *Neospathidium longinucleatum* (back; see Fig. 12.9j–l in Chapter 12)

In memory of Wilhelm Foissner (1948–2020)

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Preface, authorship, acknowledgements, and funding

The spathidiids have been one of several favorite ciliate groups of Wilhelm Foissner. In 2001, W. Foissner started a revision of this large group of haptorids. During processing his huge archive after his sudden death in 2020, I found a well-advanced manuscript dealing with several spathidiid genera. In order to prevent this manuscript from being forgotten, I have decided to publish it in my monographic series on ciliates.

W. Foissner collected most samples, made the in vivo observations, the preparations, many morphometries, and wrote text. K. Xu made morphometries and illustrations, compiled the plates, and wrote text. I updated the text of the raw manuscript, organized the deposition of the slides in the Biology Centre of the Upper Austrian Museum in Linz, wrote the front matter, the general introduction, the material and method section including the summary of taxa (Chapter 1), the brief introduction to the spathidiids (Chapter 2), the chapter on *Neocultellothrix* Foissner nov. gen. (Chapter 13), and the back matter (index). Further, I made the layout and produced the final PDF.

The help of the following persons must be acknowledged: Sabine Agatha, Remigius Geiser, Eva Herzog, Wolf-Dietrich Krautgartner, Brigitte Moser, Birgit Peukert, Fritz Seyrl, and Andreas Zankl. Colleagues who provided samples are acknowledged in the individual species descriptions. I also want to thank Magdalini Christodoulou and Alexandra Aberham at the Biology Centre of the Upper Austrian Museum in Linz for help with the transfer of the Foissner archive from Salzburg to Linz.

Wilhelm Foissner, Kuidong Xu, and co-workers involved in this project got financial support by the Austrian Science Fund FWF (Project P15017-B06, "Monographie der Familie Spathidiidae (Ciliophora)"). I wish to thank Ilse Foissner who generously privately financed my work on this book.

Salzburg
January 2025

Helmut Berger (Publisher)
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Abstract

Foissner W., Xu K. & Berger H. (Eds) (2025): Revision of some spathidiid genera (Alveolata, Ciliophora, Spathidiida). – Ser. Monogr. Cilioph. 6: i–xv, 1–465.

This book deals with some spathidiid taxa. The following genera are treated and established, respectively: *Apospathidium* Foissner et al., 2002; *Centrospathidium* nov. gen.; *Epispathidium* Foissner, 1984; *Latispathidium* Foissner et al., 2005; *Schmidingerophrya* nov. gen.; *Semibryophyllum* nov. gen.; *Semispathidium* Foissner et al., 2002; *Supraspathidium* Foissner & Didier, 1981; *Pharyngospathidium* nov. gen. (type genus of Pharyngospathidiidae nov. fam.); *Neospathidium* nov. gen.; *Neocultellothrix* Foissner nov. gen. The latter genus "replaces" *Cultellothrix* Foissner, 2003, an unavailable genus because no holotype was fixed for the type species in the original description. In addition, 12 *Spathidium* species are reviewed, and three new species assigned to this genus are described. In total, four new subspecies, 19 new species, six new genera, and one new family are described, 13 species are transferred to other genera, and 41 known species and two subspecies are reviewed. Further, three "*Spathidium* groups" are discussed. The type slides of the new species and voucher slides of the redescribed species are documented.

Key words: Alveolata; biogeography; Ciliophora; cyst; diversity; Haptoria; monograph; morphogenesis; nomenclature; Protista; revision; soil biology; systematics; taxonomy

Chapter 6

Epispathidium Foissner, 1984 (Ciliophora, Spathidiidae), a genus where the circumoral kinety is completely separated from the somatic kineties[1]

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Abstract

In the species assigned to *Epispathidium* Foissner, 1984, the circumoral kinety is completely separated from the anterior end of the somatic kineties. Further, the anterior end of the somatic kineties has a distinctly condensed ciliature which is curved ventrally on the left side of the body and dorsally on the right side. One new species, *Epispathidium salsum* nov. spec., discovered in highly saline soil from the Pacific Coast of the USA, is described. Three species (*Epispathidium regium*, type species by original designation; *Epispathidium papilliferum*, *Epispathidium securiforme*) are revised in detail while three species (*Epispathidium amphoriforme*, *Epispathidium ascendens*, *Epispathidium terricola*) are briefly reviewed. A key to the species is provided. *Spathidium macrostomum* Wilbert, 1995 is a junior primary homonym of *Spathidium macrostomum* Wang & Nie, 1933.; it is replaced by *Spathidium canadense* Wilbert nom. nov. *Spathidium polymorphum* Wenzel, 1955 is classified as objective synonym of *Epispathidium ascendens* (Wenzel, 1955) Foissner, 1987 because both species are based on the same clone. *Epispathidium polynucleatum* Foissner et al., 2002 is no longer classified in the present genus; it was transferred to *Spathidium*: *Spathidium polynucleatum* (Foissner et al., 2002) Jang et al., 2017.

[1] This chapter should be referenced as follows: Foissner W., Xu K. & Berger H. (2025): *Epispathidium* Foissner, 1984 (Ciliophora, Spathidiidae), a genus where the circumoral kinety is completely separated from the somatic kineties. – Ser. Monogr. Cilioph. 6: 141–211.
For notes on "Material and methods", see Chapter 1 (Berger et al. 2025a).

Epispathidium Foissner, 1984[2]

1984 ***Epispathidium* nov. gen.**[3] – Foissner, Stapfia 12: 81 (original description; for diagnosis see below). Type species (by original designation): *Epispathidium regium* Foissner, 1984.
2007 ***Epispathidium*** – Foissner & Xu, Monogr. biol. 81: 13, Fig. 8i, j (characterization of the *Epispathidium* ciliature).
2007 ***Epispathidium* Foissner, 1984** – Jankowski, Phylum Ciliophora, p. 564 (generic revision of ciliates).
2008 ***Epispathidium* Foissner, 1984** – Lynn, Ciliated protozoa, p. 370 (familial revision of ciliates).
2017 ***Epispathidium*** – Jang, Vdačný, Shazib & Shin, J. nat. Hist. 51: 971 (discussion of validity of *Epispathidium*).

Nomenclature: No etymology has been provided in the original description or a later work. *Epispathidium* is a composite of the Greek prefix *epi+* (with many meanings; in present case likely "on" or "beside" or "above" of a structure or "headwards"; Werner 1972, p. 65) and the genus-group name *Spathidium* (for etymology of this name, see Chapter 2, that is, Berger et al. 2025b). The name refers to the fact that the circumoral kinety is a closed row of dikinetids and arranged "on" or "above" the anterior end of the somatic kineties. Like *Spathidium* of neuter gender (Aescht 2001, p. 282).

Diagnosis (from Foissner 1984, slightly modified): Spathidiidae with circumoral kinety completely separated from somatic kineties. Anterior end of somatic kineties with distinctly condensed ciliature, on left side curved ventrally and on right side dorsally. Oral bulge in top view elongate orthogonal, very distinctly set off from body proper, moderately strongly slanted ventrally. Row 3 of dorsal brush shortened by about 50%, composed of much less dikinetids than rows 1 and 2.

Species originally assigned: *Epispathidium regium* Foissner, 1984 (type species); *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 (original combination *Spathidium amphoriforme*); *Epispathidium papilliferum* (Kahl, 1930a) Foissner, 1984 (original combination *Spathidium papilliferum*). Foissner (1984, p. 82) mentioned "*Spathidium (Epispathidium?) bavariense* Kahl, 1930"; however, this is not a formal combination and thus *Spathidium bavariense* Kahl, 1930a (p. 384) is not a species originally included.

Species incorrectly assigned: *Epispathidium polynucleatum* Foissner, Agatha & Berger, 2002 (now *Spathidium polynucleatum* (Foissner et al., 2002) Jang et al., 2017; see Chapter 3, that is, Foissner et al. 2025).

Species now assigned (see remarks): *Epispathidium regium* Foissner, 1984 (type species); *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 (original combination *Spathidium amphoriforme*); *Epispathidium ascendens* (Wenzel, 1955) Foissner, 1987 (original combination *Spathidium ascendens*); *Epispathidium papilliferum* (Kahl, 1930a) Foissner, 1984 (original combination *Spathidium papilliferum*); *Epispathidium salsum* nov. spec.; *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (original combination *Spathidium amphoriforme* var. *securiforme*); *Epispathidium terricola* Foissner, 1987 (transferred to *Spathidium* by Jang et al. 2017, p. 971).

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[2] Note by H. Berger: The genus section and the "Brief review of other species assigned to *Epispathidium* Foissner, 1984" (p. 196) was not available in the raw manuscript of W. Foissner. These parts were written by H. Berger.

[3] Foissner (1984) provided the following diagnosis: "Spathidiidae, deren circumorale Kinete vollständig von den Somakineten getrennt ist. Anteriore Enden der Somakineten mit deutlich verdichteter Ciliatur, verlaufen wegen ihrer starken Abbiegung vorne annähernd parallel zur circumoralen Kinete und weisen links nach ventral, rechts nach dorsal. Mundwulst in Aufsicht lang orthogonal, sehr deutlich vom Körper abgesetzt, mäßig stark nach ventral geneigt. Kinete 3 der Bürste um etwa 50 % verkürzt und aus viel weniger Basalkörperpaaren aufgebaut als die Kineten 1 und 2."

Remarks: Foissner & Xu (2007, p. 13) described the *Epispathidium* ciliature as follows: "The oral kinetofragments are aligned to a continuous circumoral kinety distinctly separate from the ciliary rows. The anterior region of the somatic kineties is densely ciliated and usually so distinctly curved dorsally (right side) or ventrally (left side) that the circumoral kinety is seemingly doubled, that is, the anterior region of the ciliary rows parallels the circumoral kinety." (see also Foissner 1984, p. 67). By contrast, the *Spathidium* pattern is characterized by oral kinetofragments that are usually connected to the somatic kineties from which they originated (Foissner & Xu 2007, p. 13). *Arcuospathidium* Foissner, 1984 has, like *Epispathidium*, an isolated circumoral kinety, but the somatic kineties are directed dorsally on both sides (Foissner & Xu 2007, p. 13). Unfortunately, these morphological differences are not reflected in the phylogenetic analyses based on gene sequence data, that is, *Epispathidium* species do not form a cluster, but are distributed at various sites in spathidiid phylogenies (see Fig. 16 in Jang et al. 2017, p. 969). Thus, most *Epispathidium* species have been classified in *Spathidium* by Jang et al. (2017). Since the type species *Epispathidium regium* is not yet analysed molecular biologically a final decision about the validity of *Epispathidium* is not possible at the present state of knowledge (for further discussion, see remarks at *Epispathidium securiforme*). In the present review we assign the species according to the morphological data as discussed by Foissner (1984) and Foissner & Xu (2007). The fact that the 18S rRNA tree of the spathidiids, published by Jang et al. (2017) contains 14 genera (*Spathidium*, *Arcuospathidium*, *Lagynophrya*, *Enchelys*, *Balantidion*, *Apobryophyllum*, *Cultellothrix*, *Trachelophyllum*, *Epispathidium*, *Enchelyodon*, *Pseudoholophrya*, *Acaryophrya*, *Semispathidium*, *Protospathidium*), with *Spathidium* species irregularly distributed over the whole tree, indicates that the taxonomy and phylogenetic analysis of the spathidiids are still in the early stages of development.

The separation of the species included in *Epispathidium* in the present revision is difficult in vivo. Protargol preparations should be made (i) to check if the specimens have a continuous circumoral kinety and (ii) to count the somatic kineties.

Key to species

1 Oral bulge with 2 or 3 papillae ..... *Epispathidium papilliferum* (p. 174)
– Oral bulge with no papillae **2**
2 Many (usually more than 50) macronuclear nodules **3**
– Macronucleus not as above **4**
3 On average 41–49 (total range 36–57) somatic kineties ..... *Epispathidium regium* (p. 144)
– On average 33 (range 20–40) somatic kineties ..... *Epispathidium salsum* nov. spec. (p. 166)
4 (2) Extrusomes about 40 µm long ..... *Epispathidium terricola* (p. 196)
– Extrusomes distinctly shorter **5**
5 On average 54 ciliary rows ..... *Epispathidium securiforme* (p. 154)
– On average 19–28 (range 17–38) somatic kineties **6**
6 On average 28 (range 24–38) somatic kineties ..... *Epispathidium amphoriforme* (p. 197)

– On average 19–21 (total range 17–25) somatic kineties[4] .... *Epispathidium ascendens* (p. 202)

Epispathidium regium Foissner, 1984

(Fig. 6.1a–p, 6.2a–o, Table 6.1)

1984 ***Epispathidium regium* nov. spec.**[5] – Foissner, Stapfia 12: 82, Abb. 41a–f, Tabelle 21 (Fig. 6.1a–f; one type slide [accession number 1984/8; see Aescht 2008, p. 175, Fig. 13] with protargol-prepared specimens has been deposited in the Biology Centre of the Upper Austrian Museum in Linz [LI], see nomenclature).

Nomenclature: *Epispathidium regium* is the type species of *Epispathidium*. No derivation of the name has been provided in the original description or a later work. The species-group name *regi·us*, *-a*, *-um* (Latin adjective [m, f, n]; royal, regal; Hentschel & Wagner 1996, p. 511) refers to the overall "majestic" appearance.

On the label of the slide which contains the type material of *Epispathidium regium*, W. Foissner wrote "*Epispath. regium* (Holotyp)" (see Fig. 13 in Aescht 2008, p. 134). In the work itself no holotype sensu stricto was fixed. However, Foissner (1984, p. 8) stated that "from each new species described in this work 1–3 holotype slides[6] have been deposited in the Upper Austrian Museum in Linz (LI)". From unpublished notes we can determine that the specimen documented in Fig. 41e, f of Foissner (1984; = Fig. 6.1e, f in present work) is the holotype specimen (ICZN 1999, Articles 72.4.1.1, 73.1.2). Four voucher slides (Fig. 6.2h–o; accession numbers 2024/143, 144, 145, 146) of the population from Costa Rica (Fig. 6.1g–p, 6.2a–g) are also deposited in the Biology Centre of the Upper Austrian Museum in Linz (LI).

Improved diagnosis (based on type population from Austria and on population from Costa Rica): Body size about 180–200 × 55–65 µm in vivo. Body spatulate with oblique, oblong oral bulge about 1.0–1.6 times as long as widest trunk region. Macronucleus in scattered in approximately 50–100 nodules. Two types of extrusomes: type I rod-shaped and slightly curved, 6–12 µm long; type II rod-shaped and about 2 µm long. On average 41–49 ciliary rows, three anteriorly modified to ordinary dorsal brush occupying about 22–29% of body length.

Remarks: The original description of *Epispathidium regium* is not very detailed because only eight specimens were available for morphometry (Foissner 1984). None the less, it is obvious that the Costa Rican population (Fig. 6.1g–p, 6.2a–g) is rather similar to the Austrian specimens (Fig. 6.1a–f). The sole, distinctly deviating feature is the ratio of oral bulge length to body width, which is on average 1.1:1 in the Austrian, but 1.6:1 in the Costa Rican specimens, whose oral bulge is thus highly prominent (Fig. 6.2a, b). The number of ciliary rows (49 vs. 41) and macronucleus nodules (about 52 vs. 100), and the length (12 µm vs. 6–8 µm) of the type I extrusomes are also slightly different, indicating some biogeographic specialization. However, it would be premature to separate the populations at the present state

[4] Values from Foissner (1987, p. 233) and Jang et al. (2017, p. 945).

[5] Foissner (1984) provided the following diagnosis: "In vivo ungefähr 150–300 × 50–80 µm großes *Epispathidium* mit etwa 100 länglichen Makronucleus-Teilen und deutlich schräg nach ventral abfallendem Mundwulst."

[6] Note by H. Berger: A holotype is a single specimen, thus, only one holotype slide can exist.

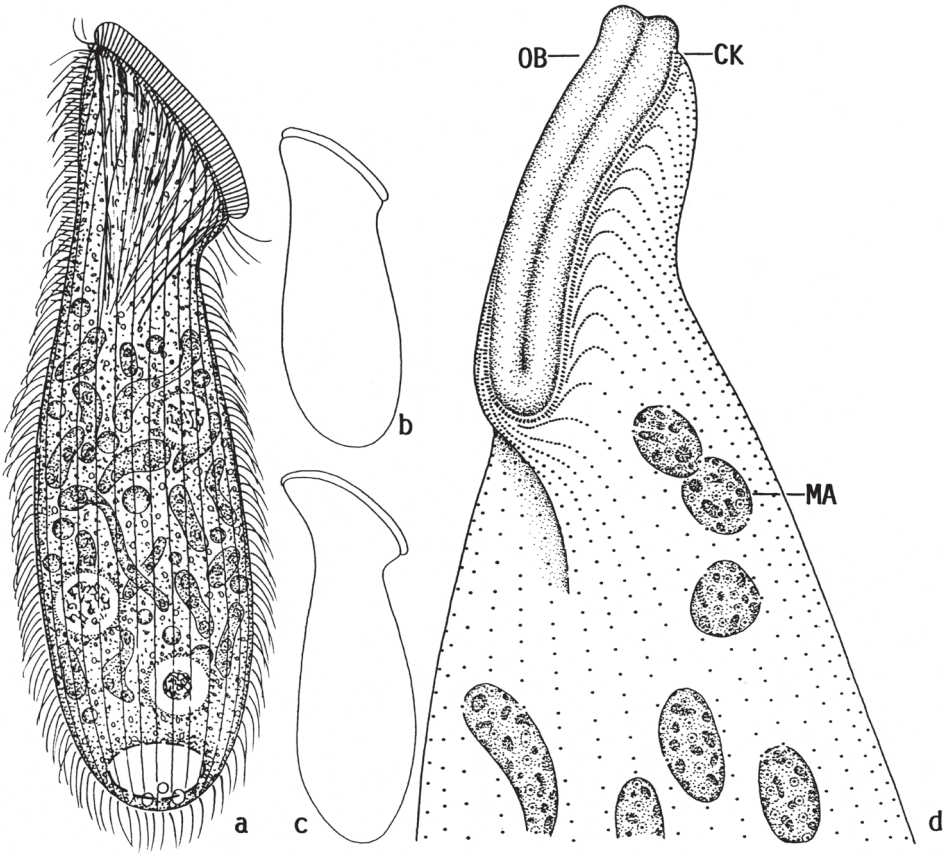


Fig. 6.1a–d *Epispathidium regium* Foissner, 1984 (from Foissner 1984. a–c, from life; d, protargol preparation). **a:** Right side view of a representative specimen of type population, 200 µm. **b, c:** Right side views of shape variants, about 130 µm, 180 µm. **d:** Ventral view of ciliary pattern in anterior body portion, length of oral bulge 42 µm. CK – circumoral kinety, MA – macronucleus nodules, OB – oral bulge.

of knowledge because all main features basically match, that is, only more or less conspicuous morphometric differences occur, which must not be over-interpreted because the Austrian data are based on only eight specimens having high variation coefficients in body and oral bulge size (Table 6.1). Thus, the diagnosis and description include both populations.

In the early 2000s, we observed two specimens each in vivo and in protargol slides from a deciduous forest in Vienna, Austria (Johannser Kogel; see Foissner et al. 2005, p. 627 for faunistic records). They largely match those from the type locality in Salzburg: length about 220 µm in vivo; body shape as in *Epispathidium amphoriforme* (p. 197) with oral bulge circa as long as trunk wide; many macronucleus nodules of highly variable size; extrusomes rod-shaped with rounded ends, slightly curved, 10 µm long, no second type noted (overlooked?); cortex with dense, plate-like cortical granulation, individual granules about 0.8 × 0.2 µm; oral bulge circa 5 µm high and 10 µm wide in vivo; about 40 ciliary rows arranged

in pronounced *Epispathidium*-pattern; anterior bristle of dorsal brush dikinetids clavate and up to 4 µm long, posterior only up to 1 µm long, both V-like spread and decreasing in length posteriorly, row 3 short but with monokinetidal bristle tail extending to near mid-body.

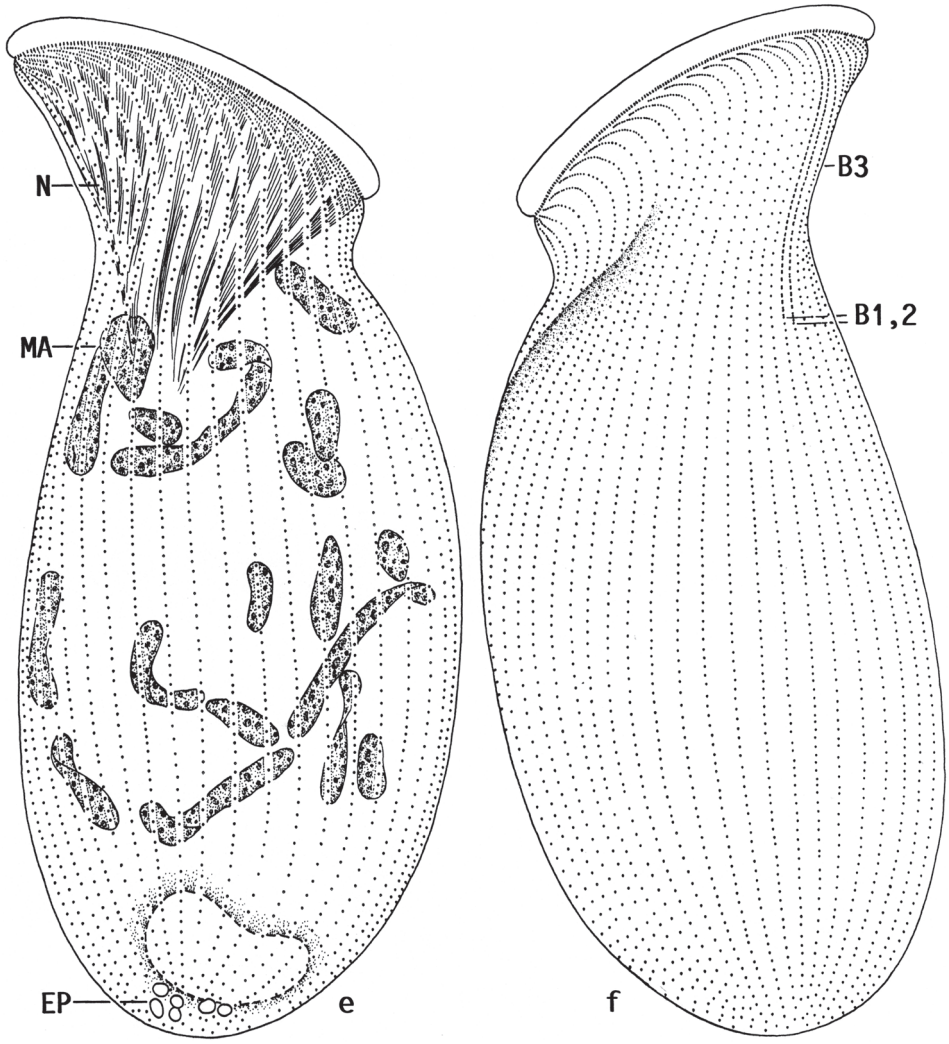
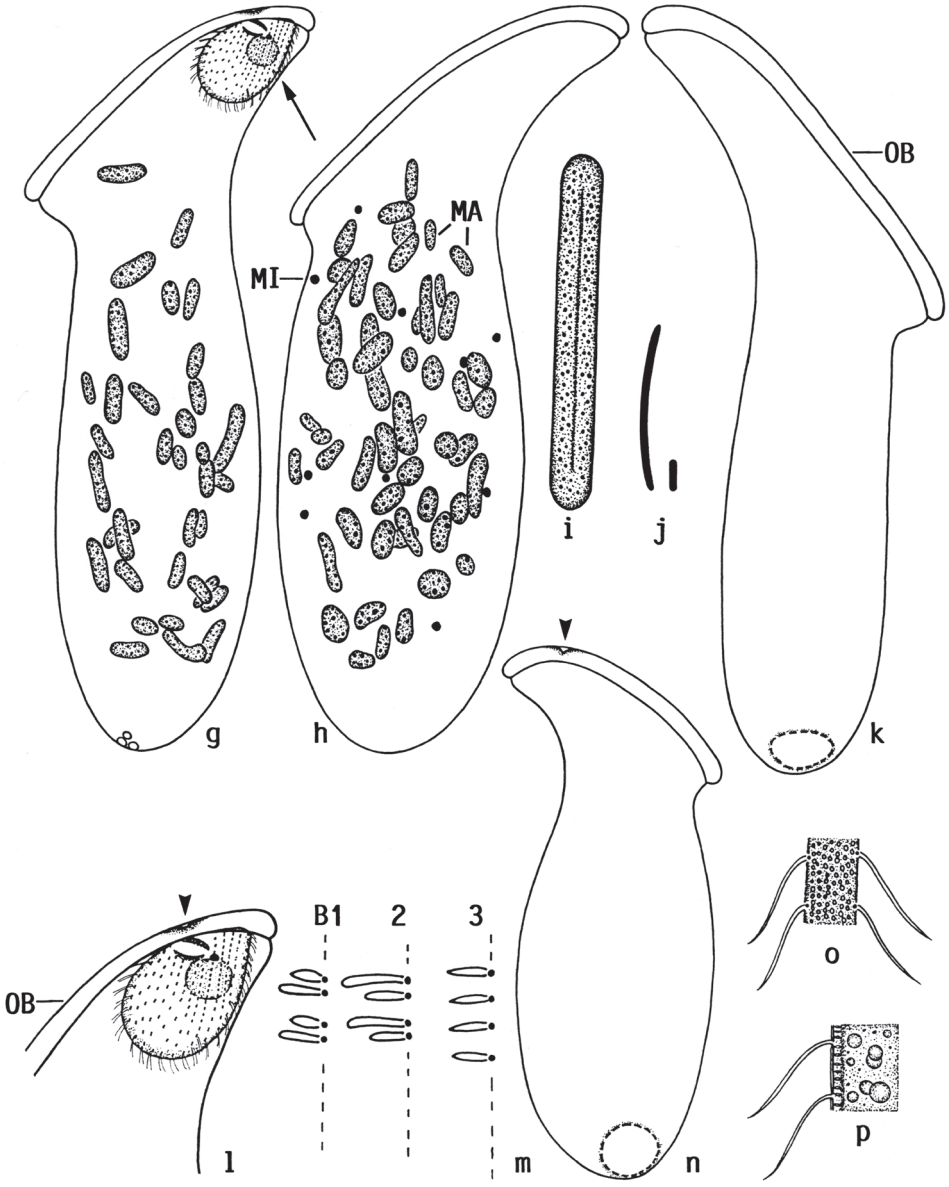


Fig. 6.1e, f *Epispathidium regium* Foissner, 1984 (from Foissner 1984. Protargol preparation). **e, f:** Right (e) and left (f) side view of holotype specimen (see nomenclature), 170 µm. B1–3 – dorsal brush rows, EP – excretory pores of contractile vacuole, MA – macronucleus nodule, N – nematodesmata.

Fig. 6.1g–p *Epispathidium regium* Foissner, 1984 (originals of specimens from Costa Rica. g, h, k, l, n, protargol preparation; i, j, m, o, p, from life). **g, l:** A specimen (205 µm; left side view) having just ingested a *Colpoda* (arrow in g), still lying under the temporary cytostome (arrowhead in l). **h, k, n:** Shape variants in left (h) and right (k, n) side views, 206 µm, 215 µm, 150 µm. Arrowhead in (n) marks temporary cytostome near dorsal bulge end. **i:** Frontal view of oral bulge. **j:** Oral bulge extrusomes, length 12 µm and 2 µm. **m:** Mid-region of dorsal brush. **o, p:** Surface view and optical section showing cortical granulation. B1–3 – dorsal brush rows, MA – macronucleus nodules, MI – micronucleus, OB – oral bulge.

Epispathidium regium is easily distinguished from the multinucleate congeners, viz., from *Epispathidium salsum* nov. spec. (body length 200 µm vs. 130 µm; >40 vs. 33 ciliary rows, extrusome length 10 µm vs. 5 µm) and *Epispathidium papilliferum* (with conspicuous oral bulge papillae), and from *Spathidium polynucleatum* (length:width ratio 3:1 vs. 6:1; 41–49 vs. 22 ciliary rows on average; see Chapter 3, that is, Foissner et al. 2025). In vivo, *Epispathidium regium* may be confused with *Spathidium canadense* Wilbert nom.

continued on p.149



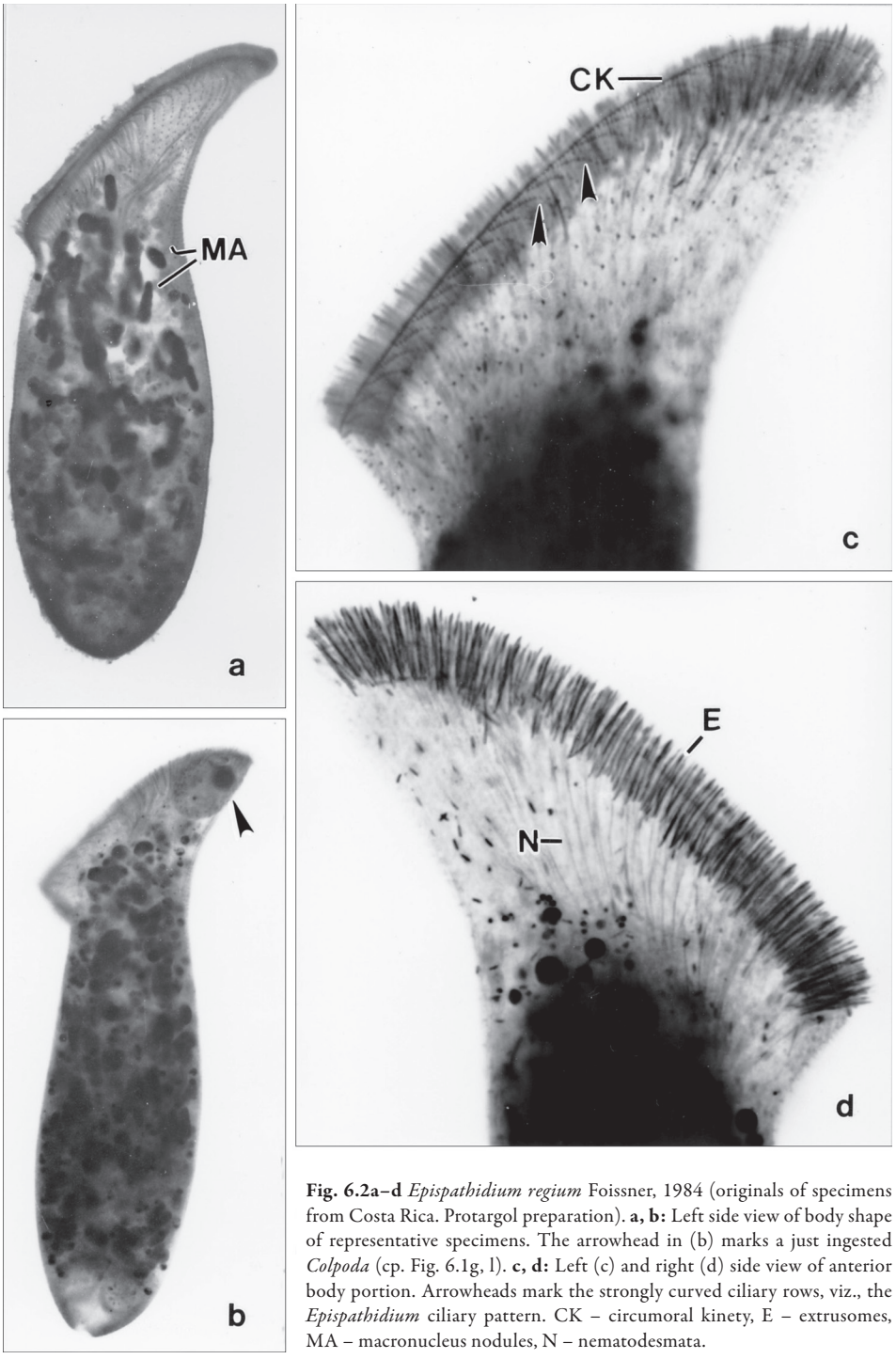


Fig. 6.2a–d *Epispathidium regium* Foissner, 1984 (originals of specimens from Costa Rica. Protargol preparation). **a, b:** Left side view of body shape of representative specimens. The arrowhead in (b) marks a just ingested *Colpoda* (cp. Fig. 6.1g, l). **c, d:** Left (c) and right (d) side view of anterior body portion. Arrowheads mark the strongly curved ciliary rows, viz., the *Epispathidium* ciliary pattern. CK – circumoral kinety, E – extrusomes, MA – macronucleus nodules, N – nematodesmata.

nov.[7] (= *Spathidium macrostomum* Wilbert, 1995; *Spathidium* ciliary pattern; body length >150 µm vs. ≤150 µm), *Spathidium seppelti etoschense* Foissner et al., 2002 (*Spathidium* ciliary pattern; body length usually >150 µm vs. ≤150 µm; 41–49 vs. 28 ciliary rows on average), and *Arcuospathidium multinucleatum* Foissner, 1999 (*Arcuospathidium* ciliary pattern; body length usually >150 µm vs. <150 µm; 41–49 vs. 15 ciliary rows on average; for revision, see Foissner & Xu 2007, p. 173). The multinucleate species of the new genera *Pharyngospathidium* and *Neospathidium* have a permanent cytostome, i.e., a distinct concavity in the oral bulge ((see Chapter 12, that is, Foissner et al. 2025a).

Description: The description is based on the Austrian type population and a population from Costa Rica studied in 1991.

Body size highly variable both in vivo and in protargol preparations (Table 6.1): 150–300 × 50–80 µm in vivo and 132–260 × 42–72 µm, on average 166 × 54 µm in protargol preparations of type population; 150–218 × 42–67 µm, on average 185 × 55 µm in protargol-impregnated Costa Rican specimens. Taking into account the in vivo measurements and some preparation shrinkage, an usual in vivo size of 180–200 × 55–65 µm and a length:width ratio of about 3:1 can be calculated. Body shape also rather variable, viz., spatulate to almost knife-like with usually distinctly narrowed neck separating the ellipsoidal trunk from the axe-shaped, strongly oblique oral (anterior) portion on average as long as (type population) or 1.6 times longer (Costa Rican specimens) than widest trunk region; hyaline oral area up to 3:1 flattened, trunk only slightly flattened and broadly rounded posteriorly (Fig. 6.1a–c, e, g, h, k, n, 6.2a, b). Macronucleus composed of about 50 (Costa Rican specimens) to 100 (type population) nodules scattered throughout trunk; nodule shape highly varies from globular to elongate ellipsoidal and irregularly curved; many small nucleoli. Micronuclei scattered between macronucleus nodules, 2–3 µm across, exact number not recognizable due to many similar-sized and impregnated cytoplasmic inclusions (Fig. 6.1a, d, e, g, h, 6.2a). Contractile vacuole in rear body end, some excretory pores in pole area. Two types of extrusomes studded in oral bulge and scattered in cytoplasm (Fig. 6.1a, i, j, 6.2c–f): type I rod-shaped, 6–8 µm long, and not impregnated (Foissner's method) in type population, while rod-shaped, slightly curved, about 12.0 × 0.8 µm in size, and strongly impregnated (Wilbert's method) in specimens from Costa Rica; type II inconspicuous, viz., rod-shaped and only about 2 µm long, likely overlooked in type specimens. Cortex very flexible, studded with colourless, about 1 µm long, ellipsoidal granules forming a plate-like structure. Cytoplasm colourless, usually packed with lipid droplets up to 5 µm across and food vacuoles containing remnants of middle-sized prey ciliates, for instance *Colpoda inflata*, which is still intact when just ingested (Fig. 6.1a, g, l, 6.2b). Glides slowly between soil particles and on microscope slides.

Somatic cilia about 12 µm long in vivo and narrowly spaced (Table 6.1), especially in anterior portion of rows, where they form, together with the circumoral cilia, a conspicuous,

[7] Note by H. Berger: *Spathidium macrostomum* Wilbert, 1995 (p. 274) is a junior primary homonym, but not a synonym of *Spathidium macrostomum* Wang & Nie, 1933 (p. 25; original spelling "*Spathidium macrostoma* Wang & Nie, 1933"; *Spathidium* is of neuter gender [Aescht 2001, p. 300], thus *macrostomum*). According to ICZN (1999, Article 57.2), the junior name is permanently invalid. Thus, we introduce the replacement name *Spathidium canadense* Wilbert nom. nov. for the species described by Wilbert (1995) from Canada. The species-group name *canadens-is, -is*, *-e* (m, f, n; living/occurring in Canada; Hentschel & Wagner 1996, p. 150) is a composite of *canad-* (for Canada) and the Latin suffix *-ensis* (Werner 1972, p. 44), referring to the location of the type locality. In future, this species has to be cited as "*Spathidium canadense* Wilbert in Foissner, Xu & Berger, 2025".

Table 6.1 Morphometric data on *Epispathidium regium* from Austrian type locality (upper line, from Foissner 1984) and from a population from Costa Rica (lower line, original data)[a]

Characteristic	Mean	M	SD	SE	CV	Min	Max	n
Body, length	166.4	160.0	41.6	14.7	25.0	132.0	260.0	8
	184.8	185.0	20.9	4.6	11.3	150.0	218.0	21
Body, width	54.1	50.5	10.4	3.7	19.2	42.0	72.0	8
	55.3	56.0	6.3	1.4	11.4	42.0	67.0	21
Body length:width, ratio	3.1	–	–	–	–	–	–	–
	3.4	3.3	0.5	0.1	14.4	2.7	4.8	21
Oral bulge, length	57.1	57.5	10.2	3.6	17.9	42.0	72.0	8
	90.0	90.0	14.8	3.2	16.4	62.0	118.0	21
Oral bulge length:body width, ratio	1.1	–	–	–	–	–	–	8
	1.6	1.6	0.3	0.1	18.5	1.3	2.6	21
Oral bulge, height[b]	4.5	4.0	0.9	0.2	19.2	3.0	7.0	21
Circumoral kinety to last dikinetid of brush row 1, distance	37.3	39.0	5.0	1.9	13.5	28.0	42.0	7
	52.9	50.0	5.9	1.8	11.2	45.0	61.0	11
Circumoral kinety to last dikinetid of brush row 2, distance	37.3	39.0	5.0	1.9	13.5	28.0	42.0	7
	52.8	52.0	5.6	1.7	10.6	46.0	60.0	11
Circumoral kinety to last dikinetid of brush row 3, distance	15.3	14.0	3.7	1.4	24.1	10.0	20.0	7
	15.2	12.0	4.8	1.5	31.7	10.0	24.0	11
Anterior body end to first macronucleus nodule, distance	–	–	–	–	–	–	–	–
	39.2	38.0	14.9	3.2	37.9	18.0	70.0	21
Macronucleus figure, length[b]	127.5	130.0	14.0	3.1	11.0	105.0	153.0	21
Macronucleus nodules, length	16.4	15.5	8.3	2.9	50.3	8.0	35.0	8
	12.8	11.0	5.9	1.3	46.3	5.0	33.0	21
Macronucleus nodules, width	4.2	4.1	0.4	0.1	8.4	4.0	5.0	8
	4.9	5.0	1.2	0.3	24.0	3.0	7.0	21
Macronucleus nodules, number	about 100							
	51.7	52.0	8.1	1.9	15.7	38.0	65.0	19
Somatic kineties, number	41.4	41.0	3.5	1.2	8.4	36.0	46.0	8
	48.7	48.0	3.6	0.8	7.4	42.0	57.0	21
Basal bodies in a right-side kinety, number	86.3	97.0	33.7	11.9	39.0	80.0	115.0	8
	–	–	–	–	–	–	–	–
Dorsal brush rows, number	3.0	3.0	0.0	0.0	0.0	3.0	3.0	8
	3.0	3.0	0.0	0.0	0.0	3.0	3.0	21
Dikinetids in brush row 1, number[b]	52.0	52.0	8.1	2.4	15.5	39.0	65.0	11
Dikinetids in brush row 2, number[b]	50.0	50.0	5.7	1.7	11.5	40.0	60.0	11
Dikinetids in brush row 3, number[b]	15.4	15.0	4.6	1.4	30.0	10.0	28.0	11

[a] Data based on mounted, protargol-prepared (Foissner's method, upper line; Wilbert's method, lower line), and randomly selected specimens from non-flooded Petri dish cultures. Measurements in µm. CV – coefficient of variation in %, M – median, Max – maximum, Mean – arithmetic mean, Min – minimum, n – number of individuals investigated, SD – standard deviation, SE – standard error of arithmetic mean.

[b] Data refer to population from Costa Rica.

metachronically beating ciliary corona (Fig. 6.1a, d–f, 6.2e, f). On average 41 (type population) to 49 (Costa Rican specimens) bipolar, rather closely spaced ciliary rows arranged in typical *Epispathidium* pattern anteriorly; rows of Costa Rican specimens anteriorly

continued on p. 152

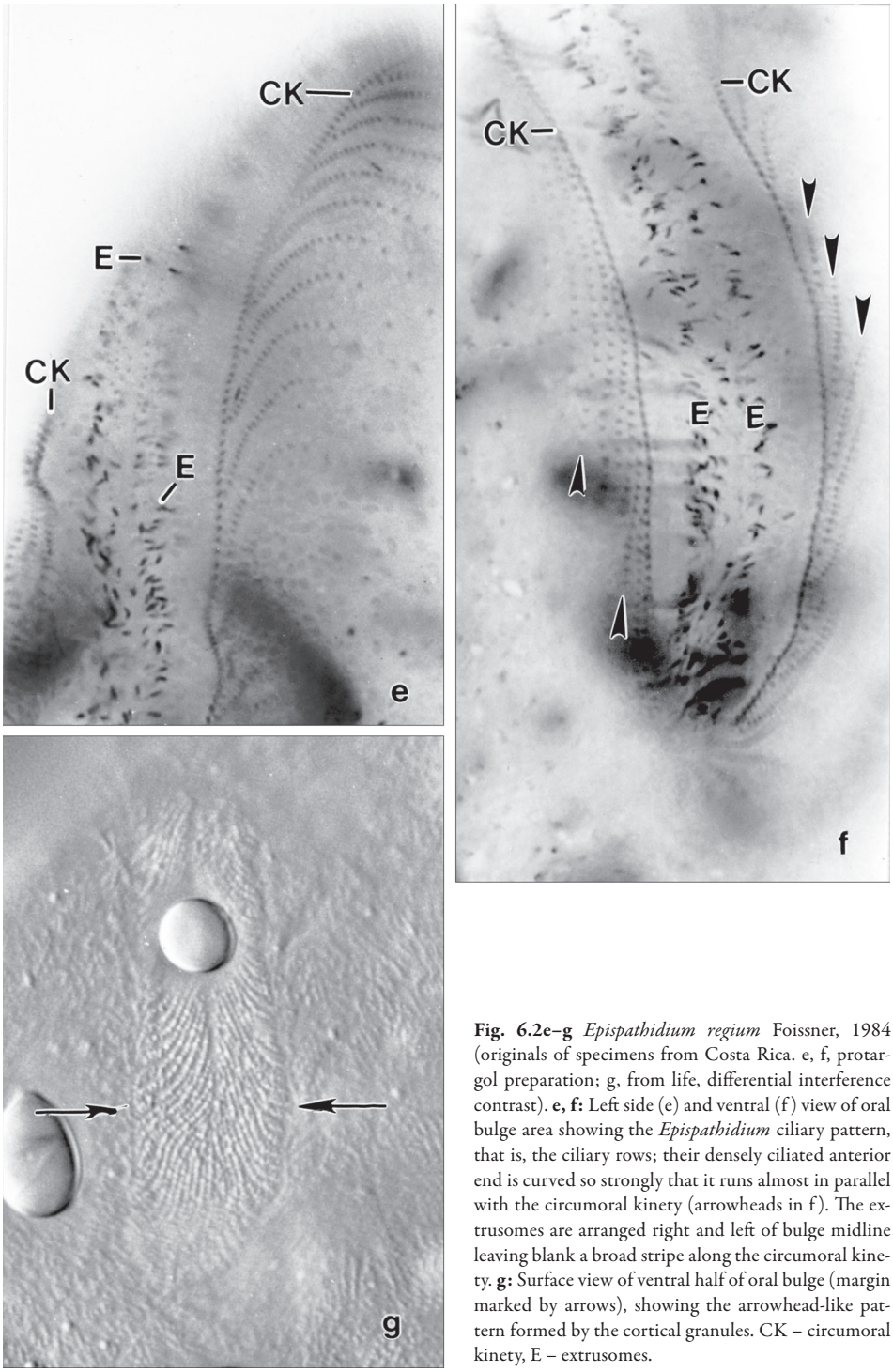


Fig. 6.2e–g *Epispathidium regium* Foissner, 1984 (originals of specimens from Costa Rica. e, f, protargol preparation; g, from life, differential interference contrast). **e, f:** Left side (e) and ventral (f) view of oral bulge area showing the *Epispathidium* ciliary pattern, that is, the ciliary rows; their densely ciliated anterior end is curved so strongly that it runs almost in parallel with the circumoral kinety (arrowheads in f). The extrusomes are arranged right and left of bulge midline leaving blank a broad stripe along the circumoral kinety. **g:** Surface view of ventral half of oral bulge (margin marked by arrows), showing the arrowhead-like pattern formed by the cortical granules. CK – circumoral kinety, E – extrusomes.

Fig. 6.2h–k *Epispathidium regium* Foissner, 1984 (originals. Protargol slides). **h–k:** Slides (h, j) and protocols (i, k) of voucher specimens (V) and voucher specimens drawn (VD) of population from Costa Rica. Accession numbers (LI): 2024/143, 144.

often with small irregularities, such as minute breaks and/or supernumerary kinetids outside rows. Dorsal brush composed of dikinetids and three-rowed, of ordinary distinctness, that is, occupies 22% (type population) to 29% (Costa Rican specimens) of body length; bristles in vivo up to 4 µm long and occasionally obliquely arranged in Costa Rican specimens; all brush rows commence with some ordinary cilia, rows 1 and 2 end at nearly same level and continue with ordinary cilia posteriorly, row 3 shorter by 60–71% than rows 1 and 2, but with a monokinetidal tail of about 2 µm long bristles extending to posterior body end (Fig. 6.1a, f). Details (shape, length, etc.) of bristles difficult to observe due to their narrow spacing, the strong flexibility of the body, and the incessant movement of the cells; possibly as shown in Figure 6.1m.

Oral bulge slanted by 30° to 60°, conspicuous because rather distinctly set off from body proper, up to 7 µm high, and about as long as (type population) or 1.6 times longer (Costa Rican specimens) than widest trunk region, both in specimens prepared with Foissner's and Wilbert's protargol protocol; surface slightly to distinctly convex in lateral view, rarely flat; elongate elliptical in frontal view (Fig. 6.1a–i, k, n; 6.2a, b, e–g). Temporary cytostome near dorsal end of bulge, distinct only in some well-impregnated and oriented Costa Rican specimens, of which one contains a just engulfed, still intact, about 30 × 20 µm-sized *Colpoda* underneath the cytostome, showing that it is the main gateway for feeding (Fig. 6.1g, l, 6.2b). Circumoral kinety of same shape as oral bulge, continuous and distinctly separate from somatic ciliary rows; composed of very narrowly spaced dikinetids each associated with a cilium, a fine fibre extending into oral bulge, and a nematodesma.

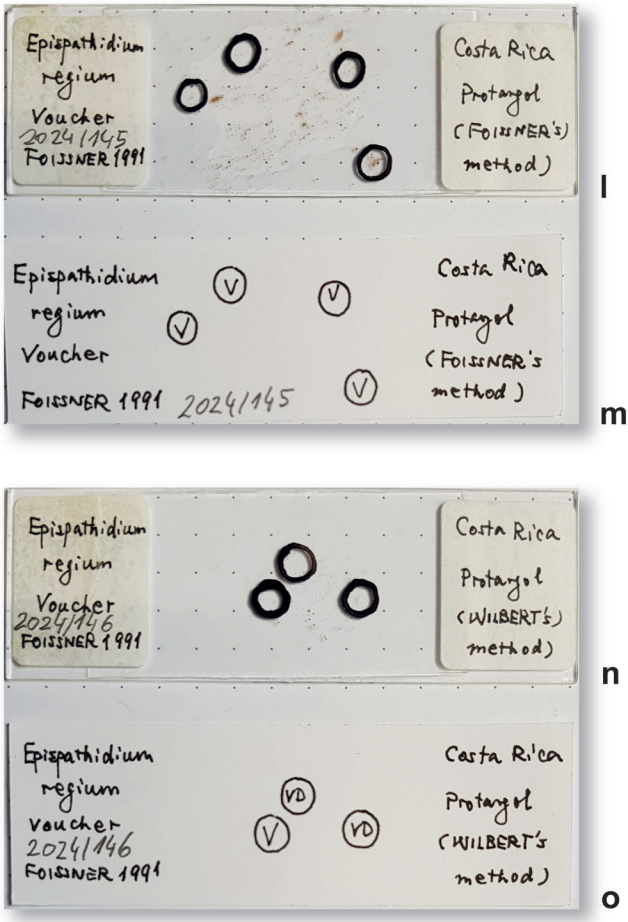


Fig. 6.2l–o *Epispathidium regium* Foissner, 1984 (originals. Protargol slides). **l–o:** Slides (l, n) and protocols (m, o) of voucher specimens (V) and voucher specimens drawn (VD) of population from Costa Rica. Accession numbers (LI): 2024/145, 146.

Oral basket moderately conspicuous in protargol preparations, because composed of more or less distinct nematodesma bundles extending to second body third (Fig. 6.1d, e, i, 6.2a–f).

Occurrence and ecology: The data below indicate that *Epispathidium regium* is a rare, euryoecius cosmopolitan leaf litter species. The big size makes it unlikely that it occurs in fine-pored mineral soil. The type locality is soil from an alder stand (about 49°07'N 13°07'E, about 1780 m above sea-level) on the Stubnerkogel (Austrian Central Alps) near the city of Bad Gastein, Salzburg, Austria (Foissner 1984, p. 5, 7, 82). The abundance was low at the type locality, a strongly acidic (pH 3.4 in 0.01M $CaCl_2$ solution), raw humus-like moder soil of an *Alnetum viridis* stand (for detailed description of site, see "Taxotop D" in Foissner & Peer 1985, p. 30).

Later, we found a few specimens each in moss from a beech trunk near the Tauernbach, a brook along the alpine street "Felber-Tauern-Straße" (Carinthia, Austria), and in litter from a mixed deciduous and coniferous forest near the University of the town of Kaiserslautern in Germany.[8] Foissner et al. (2005, p. 627) studied the soil ciliates from several natural forest stands in eastern Austria; the present species was recorded at all sites.

In Central America, *Epispathidium regium* was also rare in the non-flooded Petri dish culture and occurred in highly saline (salt content >20‰; pH 7.6) and sandy coastal soil near Punta Pirikiki (9.659°N 82.749°W), a headland in Puerto Viejo de Talamanca, about 50 km south of the town of Puerto Limón, Caribbean coast of Costa Rica. The sample com-

[8] Note by H. Berger: In the raw manuscript, W. Foissner did not provide references or more details on these sample sites. I suppose that these are unpublished observations.

prised humic sand, plant roots, and dry leaves from halophytes under coco palms. Foissner (1998, p. 203) mentioned a palaeotropic record.

Epispathidium securiforme (Kahl, 1930) Foissner, 1984

(Fig. 6.3a, b, e–n, 6.4a–l, Table 6.2)

1930 ***Spathidium amphoriforme* var. *securiforme* var. n.** – Kahl, Arch. Protistenk. 70: 380, Fig. 9o (Fig. 6.3a; original description; no type material available; see nomenclature).

1930 ***Spathidium amphoriforme* var. *securiforme* Kahl, 1930** – Kahl, Tierwelt Dtl. 18: 166, Fig. 22_{28} (Fig. 6.3b; revision of ciliates).

1943 ***Sp. amphoriforme* var. *securiforme* Kahl** – Kahl, Infusorien, p. 27, Tafel VI, Fig. 9 (review of ciliates).

1984 ***Epispathidium amphoriforme* (Greeff, 1889) nov. comb.** – Foissner, Stapfia 12: 82, Abb. 42a–f, Tabelle 21 (combination of species and subtaxa with *Epispathidium*, see nomenclature).

1987 ***Epispathidium amphoriforme* var. *securiforme* (Kahl, 1930) nov. comb.** – Foissner, Arch. Protistenk. 133: 224 (combination of variety with *Epispathidium*, see nomenclature).

Table 6.2 Morphometric data on *Epispathidium securiforme* from Brazil (original data)[a]

Characteristic	Mean	M	SD	SE	CV	Min	Max	n
Body, length	190.0	192.0	16.0	3.5	8.4	162.0	210.0	21
Body, maximum trunk width	54.3	53.0	7.6	1.7	14.0	45.0	73.0	21
Body length:trunk width, ratio	3.6	3.6	0.5	0.1	13.9	2.3	4.4	21
Oral bulge, length of cord	94.1	93.0	9.3	2.0	9.9	73.0	111.0	21
Oral bulge length:trunk width, ratio	1.8	1.9	0.3	0.1	18.0	1.2	2.2	21
Oral bulge, height	4.6	5.0	0.5	0.1	11.6	3.5	5.0	21
Circumoral kinety to last dikinetid of brush row 1, distance	59.2	58.0	6.0	1.3	10.1	50.0	78.0	21
Circumoral kinety to last dikinetid of brush row 2, distance	58.4	57.0	5.9	1.3	10.0	50.0	75.0	21
Circumoral kinety to last dikinetid of brush row 3, distance	13.3	12.0	2.2	0.5	16.5	10.0	18.0	21
Anterior body end to macronucleus, distance	82.0	83.0	16.1	3.5	19.6	53.0	125.0	21
Macronucleus figure, length	76.1	74.0	16.8	3.7	22.1	46.0	103.0	21
Macronucleus, length (spread, thus approximate)	304.3	320.0	–	–	–	180.0	430.0	21
Macronucleus, width in middle third	5.5	5.0	0.7	0.2	13.7	5.0	7.0	21
Macronucleus, number	1.0	1.0	0.0	0.0	0.0	1.0	1.0	21
Micronuclei, diameter	2.1	2.0	0.2	0.0	8.7	2.0	2.5	21
Somatic kineties, number	53.8	54.0	3.2	0.7	5.9	48.0	60.0	21
Basal bodies in a right-side kinety, number	98.4	95.0	11.4	2.5	11.5	78.0	122.0	21
Dorsal brush rows, number	3.0	3.0	0.0	0.0	0.0	3.0	3.0	21
Dikinetids in brush row 1, number	55.0	55.0	5.3	1.1	9.6	44.0	67.0	21
Dikinetids in brush row 2, number	49.7	50.0	4.5	1.0	9.1	42.0	60.0	21
Dikinetids in brush row 3, number	13.0	13.0	2.9	0.6	22.6	8.0	19.0	21

[a] Data based on mounted, protargol-prepared (Foissner's method), and randomly selected specimens from a non-flooded Petri dish culture. Measurements in μm. CV – coefficient of variation in %, M – median, Max – maximum, Mean – arithmetic mean, Min – minimum, n – number of individuals investigated, SD – standard deviation, SE – standard error of arithmetic mean.

2017 ***Spathidium securiforme* Kahl, 1930 stat. nov.**[9] – Jang, Vdačný, Shazib & Shin, J. nat. Hist. 51: 961, Fig. 12a–e (description of Korean population from life, see nomenclature and remarks).

Nomenclature: No derivation of the name has been provided in the original description or a later work. The species-group name *securiform·is*, *-is*, *-e* (axe-shaped) is a composite of *securis* (Latin noun, the axe, the hatchet; Hentschel & Wagner 1996, p. 530), the thematic vowel *·i-*, and *-form·is*, *-is*, *-e* (Latin adjective [m, f, n]; -shaped; see Hentschel & Wagner 1996, p. 274 at *glómeriformis*), and obviously refers to the large, convex oral portion.

No permanent preparations are available from the type population from Austria (Kahl 1930a). We do not neotypify this species via the Brazilian population described in the present work because conspecificity is not beyond reasonable doubt and detailed data on the European population are lacking. In addition, the sample sites are very far away (Austria vs. Brazil; ICZN 1999, Article 75.3.6). We deposit three voucher slides (Fig. 6.4g–l; accession numbers 2024/147, 148, 149) with protargol-prepared specimens from the population from Brazil in the Biology Centre of the Upper Austrian Museum in Linz (LI).

Kahl (1930a, b) treated the present taxon as variety of *Spathidium amphoriforme* Greeff, 1889 (p. 131).[10] According to ICZN (1999, Article 45.6.4), the rank of *Spathidium am-*

[9] Note by H. Berger: Jang et al. (2017) mentioned Kahl (1930b) as original description, which is, however, incorrect. Correct is Kahl (1930a), a description which is much more comprehensive.

[10] Note by H. Berger: Kahl (1930a) described two varieties (*Spathidium amphoriforme rectitoratum* and *Spathidium amphoriforme securiforme*) deviating more or less distinctly from the stem form ("Stammform"), that is, Kahl accepted

continued on p. 157

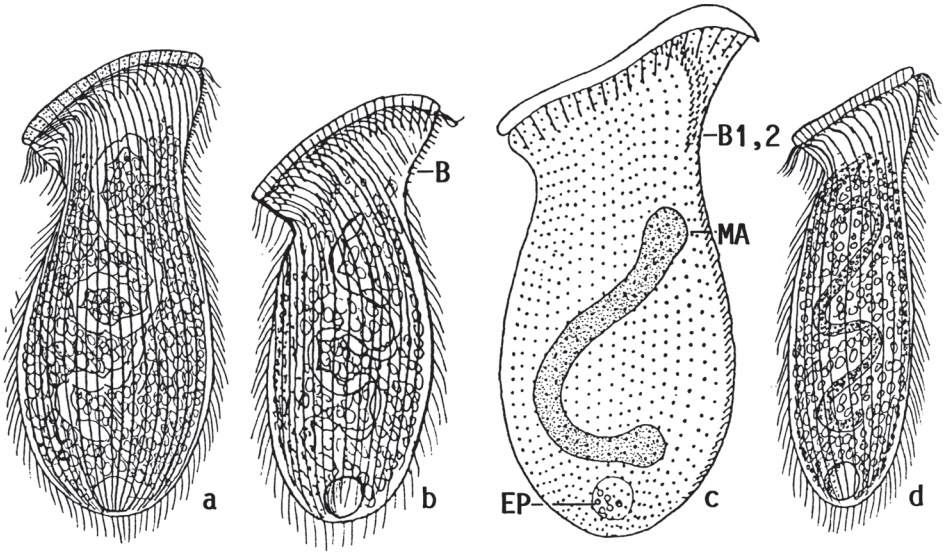


Fig. 6.3a–c *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (*Spathidium amphoriforme securiforme*; a, from Kahl 1930a; b, from Kahl 1930b; c, from Gellért 1956. a, b, from life; c, composite from opal-blue prepared specimens). **a–c:** Left side views, 200–300 µm, 250 µm, 90–100 µm. Note that Gellért's figure likely shows a *Pharyngospathidium longichilum amphoriforme* specimen.
Fig. 6.3d *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 (*Spathidium amphoriforme rectitoratum* from Kahl 1930a. From life). Left side view, 200 µm (Kahl 1930b, p. 161).

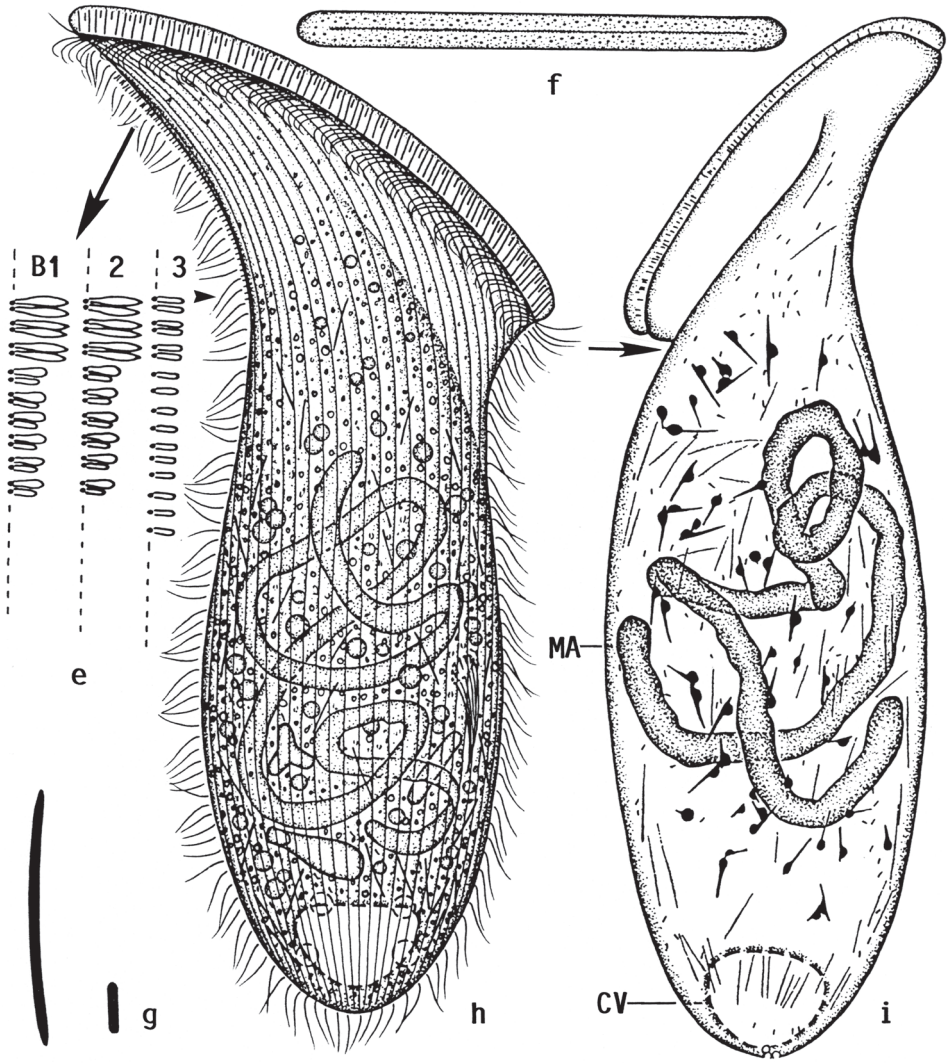
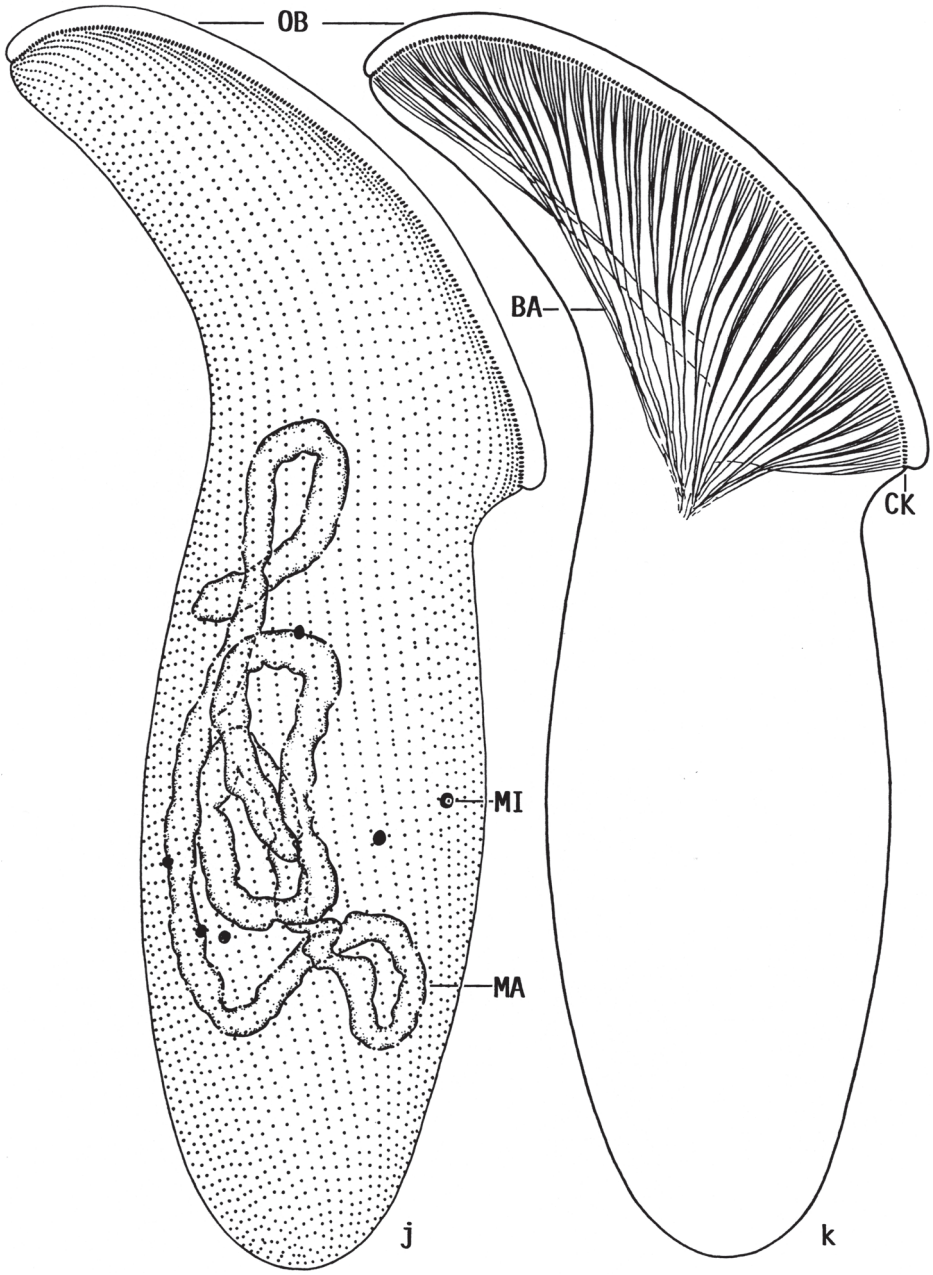


Fig. 6.3e–i *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals of Brazilian specimens. e–h, from life; i, protargol preparation). **e:** Posterior region of dorsal brush, bristles up to 5 µm long. **f:** Frontal view of oral bulge studded with type I and type II extrusomes. **g:** Type I (length 13 µm) and type II (4 µm) oral bulge extrusomes. **h:** Right side view of a representative specimen, 220 µm. Note the oral bulge which is almost twice as long as the widest trunk region, while about as long as trunk wide in the European population (Fig. 6.3a, b), a rather conspicuous difference indicating that the Brazilian population might be a different (sub)species. Arrowhead marks end of monokinetidal bristle tail of brush row 3, shown at higher magnification in (e). **i:** Dorsolateral view showing the right angle (arrow) between oral region and trunk, a feature emphasized by Kahl (1930a, b); 208 µm. Three types of extrusomes impregnated in the cytoplasm: long rods, similar to those found in the oral bulge; fusiform rods, likely developmental stages of the long rods; and very short rods, similar to those found in the oral bulge (however, these short rods could be the toxin-containing portion of developing, long type I extrusomes). B1–3 – dorsal brush rows, CV – contractile vacuole, MA – macronucleus.

Fig. 6.3j, k *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals of Brazilian specimens. Protargol preparation). **j, k:** Ciliary pattern of right side (j) and oral apparatus (k) of main Brazilian voucher specimen, 205 µm. BA – oral basket, CK – circumoral kinety, MA – macronucleus, MI – micronucleus, OB – oral bulge. →

phoriforme securiforme in Kahl (1930a) is subspecific because published before 1961 and the author used the term variety. In addition, Kahl (1930a, b) did not give the taxon expressly

three varieties within *Spathidium amphoriforme*. Interestingly, the full name of the stem form (*Spathidium amphoriforme amphoriforme* Greeff, 1889) was never mentioned. For characterization of varieties in Kahl (1930a), see Table 6.6.



infrasubspecific rank. Jang et al. (2017) raised the subspecies *Spathidium amphoriforme securiforme* to species rank. This act does not change the authorship, that is, the correct name in *Spathidium* is *Spathidium securiforme* Kahl, 1930a (ICZN 1999, Article 50.3.1).

Foissner (1984) established *Epispathidium* and transferred *Spathidium amphoriforme* to this genus. Since Foissner (1984) did not remove the two varieties established by Kahl (1930a) from *Spathidium amphoriforme*, we have to assume that the varieties have also been transferred to *Epispathidium* by Foissner (1984). Consequently, the separate transfer of *Spathidium amphoriforme securiforme* Kahl, 1930a and *Spathidium amphoriforme rectitoratum* Kahl, 1930a to *Epispathidium* by Foissner (1987a, p. 224; see list above) is superfluous. Please note that *Epispathidium amphoriforme* sensu Foissner (1984) is classified at *Epispathidium amphoriforme* (p. 197), that is, the work by Foissner (1984) is listed in the synonymy above only because of the transfer of the species and its subtaxa to *Epispathidium*.

Improved diagnosis (comprising data from type population [Kahl 1930a] and Brazilian population [original observations]): Body size about 220 × 60 µm in vivo. Body shape spatulate with axe-shaped, strongly oblique anterior (oral) portion on average 1.0–1.8 times longer than widest trunk region. Macronucleus long and tortuous. Two types of extrusomes: type I rod-shaped and slightly curved, about 13 *µm long*; type II rod-shaped and about 3 *µm long*. On average 54 ciliary rows, 3 of them anteriorly differentiated to a moderately distinct dorsal brush occupying about 31% of body length; brush rows subapically curved, wider spaced, and with obliquely arranged dikinetids.

Remarks: In the light of the insufficient and confusing data from Greeff (1889, p. 131) and Penard (1922, p. 24), Kahl (1930a) redescribed *Spathidium amphoriforme* from moss and, concomitantly, established, beside the nominotypical variety (see nomenclature above), two new varieties, namely *Spathidium amphoriforme rectitoratum* and *Spathidium amphoriforme securiforme*. *Spathidium amphoriforme rectitoratum* differs from *Spathidium amphoriforme securiforme* mainly by the straight (Fig. 6.3d) vs. convex (Fig. 6.3a, b) oral bulge, while the latter has much more ciliary rows (up to 50 vs. 24–28) and longer extrusomes (11–15 µm vs. 5 µm) (Kahl 1930a, b; for comparison of varieties as characterized by Kahl 1930a, see Table 6.6). Wenzel (1953, p. 78) did not accept the varieties because he observed transitions, however, without providing quantitative data.

Based on the investigation of an Austrian population from mixed forest soil, Foissner (1984) transferred *Spathidium amphoriforme* to *Epispathidium*, but he did not discuss the two new varieties established by Kahl (1930a) and briefly reviewed by Kahl (1930b) in detail. His population agreed with the nominotypical variety *Spathidium amphoriforme amphoriforme* Greeff, 1889 ("Stammform") redescribed by Penard (1922) and Kahl (1930a, b).

Recent, unpublished investigations on an *Epispathidium amphoriforme* population from another forest soil from Austria shows two forms with respect to the oral bulge, viz., one in which it is slightly to distinctly convex, as usual for the nominotypical variety, and another in which it is straight, as described by Kahl (1930a, b) for the variety *Spathidium amphoriforme rectitoratum*. However, there are many transitions, and both forms match perfectly in all other important features (data not shown). Thus, and because both types occur at the same site (Kahl 1930a) and even in the same sample (our data), we agree with the synonymy of *Spathidium amphoriforme amphoriforme* and *Spathidium amphoriforme rectitoratum*

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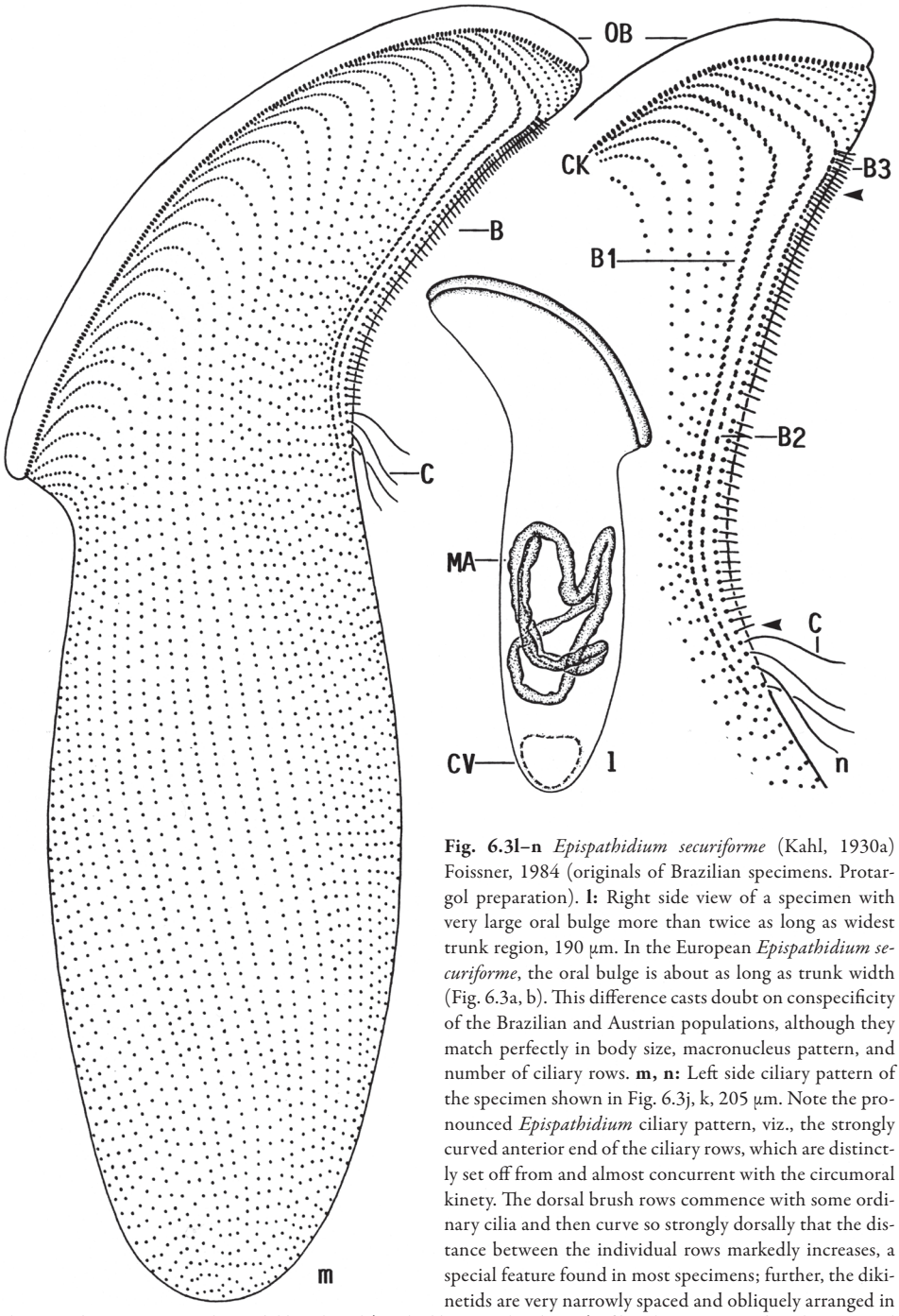

Fig. 6.3l–n *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals of Brazilian specimens. Protargol preparation). **l:** Right side view of a specimen with very large oral bulge more than twice as long as widest trunk region, 190 µm. In the European *Epispathidium securiforme*, the oral bulge is about as long as trunk width (Fig. 6.3a, b). This difference casts doubt on conspecificity of the Brazilian and Austrian populations, although they match perfectly in body size, macronucleus pattern, and number of ciliary rows. **m, n:** Left side ciliary pattern of the specimen shown in Fig. 6.3j, k, 205 µm. Note the pronounced *Epispathidium* ciliary pattern, viz., the strongly curved anterior end of the ciliary rows, which are distinctly set off from and almost concurrent with the circumoral kinety. The dorsal brush rows commence with some ordinary cilia and then curve so strongly dorsally that the distance between the individual rows markedly increases, a special feature found in most specimens; further, the dikinetids are very narrowly spaced and obliquely arranged in the curved area. The monokinetidal bristle tail (marked by arrowheads in n) of brush row 3 extends only to second body third. B – dorsal brush, B1–3 – dorsal brush rows, C – ordinary, somatic cilia, CK – circumoral kinety, CV – contractile vacuole, MA – macronucleus, OB – oral bulge.

proposed by Wenzel (1953, p. 79).[11] By contrast, Vďačný et al. (2014, p. 95) and Jang et al. (2017, p. 959) classified *Spathidium amphoriforme rectitoratum* Kahl, 1930a as distinct species in the genus *Spathidium* (*Spathidium rectitoratum* Kahl, 1930a; here preliminary classified as synonym of *Epispathidium amphoriforme*, p. 197).

The Brazilian population described in the present work largely corresponds with the variety *Spathidium amphoriforme securiforme*, especially in body and extrusome size, the macronucleus pattern and, emphatically, the high number of ciliary rows. However, there is also a difference, viz., the ratio of oral bulge length to trunk width, which is on average 1.8:1 in the Brazilian population, but only about 1:1 in Kahl's (1930a) specimens (cp. Fig. 6.3a and 6.3h); in the illustration provided by Kahl (1930b) the ratio is in between, namely about 1.3:1 (Fig. 6.3b). It is this difference that makes the Brazilian specimens looking rather dissimilar to the European ones. However, the feature is highly variable in the Brazilian population (1.2–2.2:1; Table 6.2), and detailed data on the European populations of *Epispathidium amphoriforme securiforme* are lacking. Thus, the most practicable way is, at present, to consider both populations as belonging to the same species, and to rank the subspecies *Epispathidium amphoriforme securiforme* as a distinct species in *Epispathidium*, as already proposed by Jang et al. (2017, p. 964). We do not fix the Brazilian population as neotype of *Epispathidium securiforme* because conspecificity is not beyond reasonable doubt and, more important, the two sample sites are very far away (see nomenclature).

Epispathidium securiforme differs from *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 (for brief review, see p. 197), which is likely the nearest relative, by the larger size (180–250 μm vs. 90–150 μm), the longer extrusomes (12–13 μm vs. 5–7 μm), and the much higher number of ciliary rows (on average 54 vs. 28; data from Table 6.2 and from Foissner 1984, p. 149). It differs from *Epispathidium regium* (p. 144) mainly by the macronucleus pattern (a long and tortuous strand vs. many scattered nodules). Among the Pharyngospathidiidae, only *Pharyngospathidium longichilum amphoriforme* resembles *Epispathidium securiforme*. Indeed, these two species differ only by the macronucleus (long vs. short strand) and the cytopharynx (permanent vs. temporary) (see Chapter 12, that is, Foissner et al. 2025a).

Jang et al. (2017) described a Korean population of the present species. However, since they failed to make protargol preparations, their data, which are based only on in vivo observations, should not be over-interpreted. On the basis of their phylogenetic analyses based on gene sequence data, Jang et al. (2017, p. 971) transferred all *Epispathidium* species to *Spathidium*, except for the type species (*Epispathidium regium*) for which no gene sequence data are available, that is, they did not formally classify *Epispathidium* as junior synonym of *Spathidium*. We preliminary accept the morphology-based separation of *Spathidium* (circumoral kinety composed of anterior ends of somatic kineties and thus not isolated) and *Epispathidium* (isolated circumoral kinety present) introduced by Foissner (1984, p. 67; see also Foissner & Xu 2007, p. 13) because neither *Spathidium hyalinum* Dujardin, 1841 (type species of *Spathidium*) nor *Epispathidium regium* Foissner, 1984 (type species of *Epispathidium*) have been analyzed in great detail.[12] Since the type species defines a genus (ICZN

[11] However, note that we do not agree with the synonymy of the nominotypical variety and *Spathidium amphoriforme securiforme*, likewise proposed by Wenzel (1953).

[12] Note by H. Berger: Dujardin (1841, p. 457) established *Spathidium* with *Spathidium hyalinum* Dujardin, 1841 as type species by monotypy. Dujardin (1841, p. 458) himself discussed that his species is likely identical with

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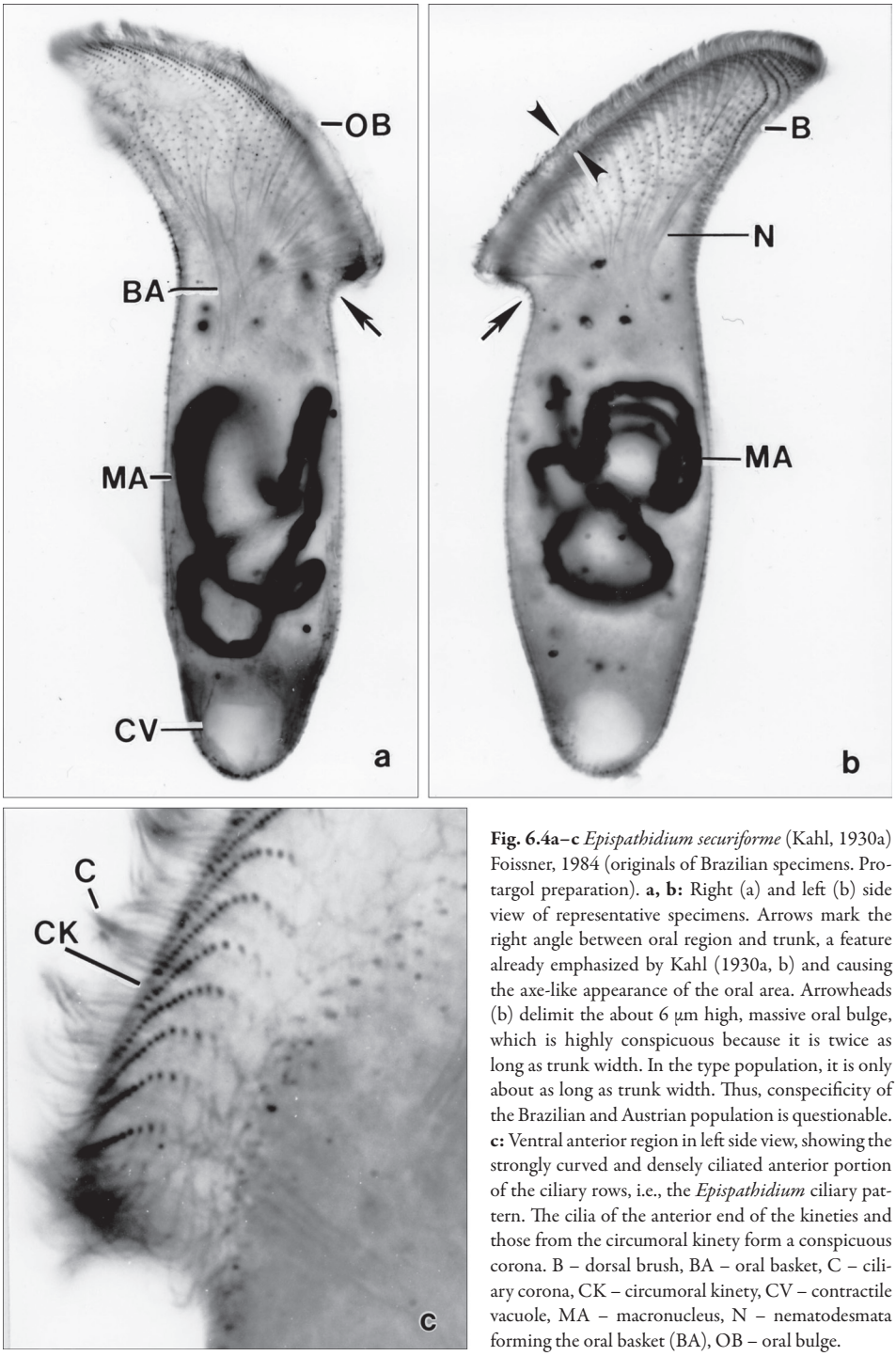

Fig. 6.4a–c *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals of Brazilian specimens. Protargol preparation). **a, b:** Right (a) and left (b) side view of representative specimens. Arrows mark the right angle between oral region and trunk, a feature already emphasized by Kahl (1930a, b) and causing the axe-like appearance of the oral area. Arrowheads (b) delimit the about 6 µm high, massive oral bulge, which is highly conspicuous because it is twice as long as trunk width. In the type population, it is only about as long as trunk width. Thus, conspecificity of the Brazilian and Austrian population is questionable. **c:** Ventral anterior region in left side view, showing the strongly curved and densely ciliated anterior portion of the ciliary rows, i.e., the *Epispathidium* ciliary pattern. The cilia of the anterior end of the kineties and those from the circumoral kinety form a conspicuous corona. B – dorsal brush, BA – oral basket, C – ciliary corona, CK – circumoral kinety, CV – contractile vacuole, MA – macronucleus, N – nematodesmata forming the oral basket (BA), OB – oral bulge.

1999, Article 67.1), a final decision on a synonymy of these two genera is not possible at the present state of knowledge. More detailed studies on reliably identified populations are needed to get a better estimation of the phylogenetic relationships of these taxa.

Gellért (1956, p. 83) redescribed the variety *Spathidium amphoriforme securiforme* from Hungarian lichens and mosses (Fig. 6.3c), but his population is very likely identical with *Pharyngospathidium longichilum amphoriforme* nov. subspec. (see Chapter 12, that is, Foissner et al. 2025a).

Description of type population from Austria (from Kahl 1930a, b): Body length 200–300 µm in vivo. Body shape rather similar to *Epispathidium amphoriforme amphoriforme*, but distinctly stouter with massive oral bulge as long as body width; neck very distinct. Macronucleus long and tortuous. Contractile vacuole in rear body end. Extrusomes studded in oral bulge and scattered in cytoplasm, 11–15 µm long and thin; Kahl (1930a) observed also specimens with extrusomes only 7–8 µm long and he supposed, as the cells were rather fragile, that the 11–15 µm long extrusomes were partially exploded short extrusomes; we would interpret the shorter extrusomes as developmental stages. About 50 somatic ciliary rows (up to 25 per side) in distinct furrows. Dorsal brush inconspicuous, bristles 2.0–2.5 µm long. All other features as in nominotypical variety according to Kahl (1930a) (Fig. 6.3a, b).

Description of Brazilian population (original data; Fig. 6.3e–n, 6.4a–f; Table 6.2): Body size 180–250 × 50–80 µm in vivo, usually about 220 × 60 µm, as calculated from some in vivo measurements and the morphometric data (Table 6.2). Body shape spatulate with very conspicuous, axe-shaped, strongly oblique anterior (oral) end about 1.8 times longer than widest trunk region; length:width ratio 2.3–4.4:1 in protargol preparations, on average near 3.6:1 both in vivo and prepared specimens; distinctly flattened only in oral region (Fig. 6.3h, j, 6.4a, b). Neck very distinct due to the large oral area less sharply defined dorsally than ventrally, where it merges into trunk at almost right angles, especially when cells are viewed dorsolaterally (Fig. 6.3i, l, 6.4a, b), as emphasized also in the key of Kahl (1930b). Trunk fusiform, dorsal side distinctly longer than ventral and conspicuously projecting anteriorly, anterior (oral) body end thus strongly slanted; posterior end moderately broadly rounded. Macronucleus usually in posterior body half, very long and highly tortuous, frequently forming 4–6, rarely up to nine coils; contains many nucleoli up to 2 µm across. Many globular micronuclei about 2 µm across near to and far from macronucleus, exact number not recognizable due to many similarly sized and impregnated cytoplasmic inclusions (Fig. 6.3h–j, 6.4a, b; Table 6.2). Contractile vacuole in rear body end, several excretory pores in pole area. Two types of extrusomes studded in oral bulge and scattered in cytoplasm (Fig. 6.3g–i): type I rod-shaped and slightly curved with ends bluntly pointed, about 13 µm long in vivo and rather thin (about 0.5 µm); certain cytoplasmic developmental stages impregnate with the protargol method used, while those attached to the oral bulge never stain; type II extrusomes rod-shaped and about 3 µm long, intensely impregnate both in oral bulge and

Enchelys spathula Müller, 1773 (p. 38; now *Spathidium spathula* (Müller, 1773) Bütschli, 1889; p. 1681). Unfortunately, the taxonomic status of this species is still uncertain because a serious neotypification and gene sequence data of a reliably identified population are lacking. The "neotypification" of *Spathidium spathula* by Foissner (1984, p. 70) mentioned by Aescht (2008, p. 179) is very likely invalid (for qualifying conditions, see ICZN 1999, Article 75.3), and for the sequence HM140392 (GenBank; Kahn & Shin, unpublished, submitted 2012) no morphological data have been published.

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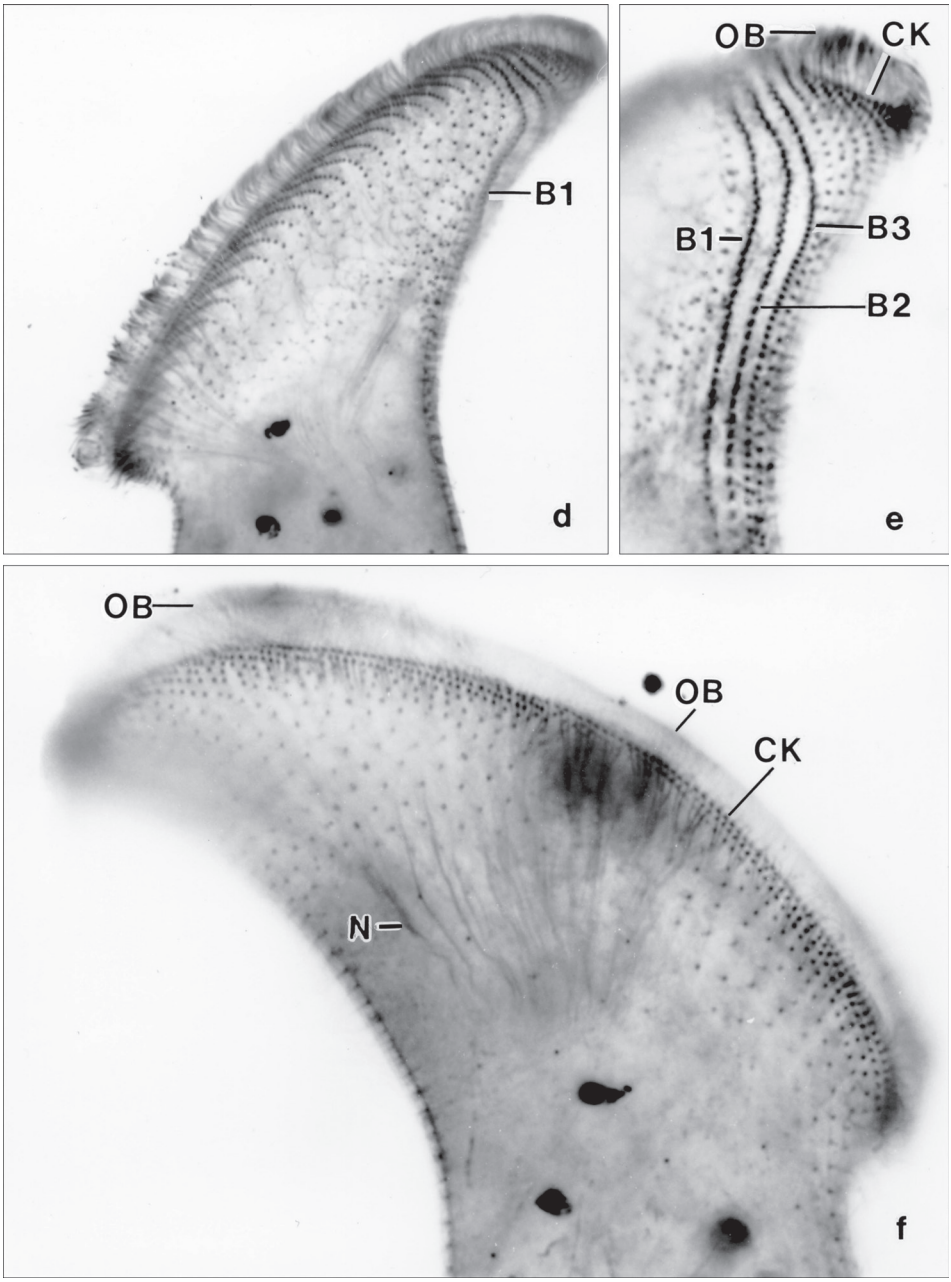


Fig. 6.4d–f *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals of Brazilian specimens. Protargol preparation). **d, f:** Left (d) and right (f) side view of oral body portion, showing the massive oral bulge and the *Epispathidium* ciliary pattern. **e:** In the curved region of the dorsal brush, the distance is enlarged between the rows and the kinetids are obliquely arranged. B1–3 – dorsal brush rows, CK – circumoral kinety, N – nematodesmata, OB – oral bulge.

cytoplasm. Cortex rather flexible. Cytoplasm colourless, often packed with lipid droplets, extrusomes, and remnants of prey, including diatoms; flagellates; resting cysts of the suctorian ciliate *Podophrya*; and other ciliates, as indicated by decaying membranelles and oral baskets. Movement without peculiarities.

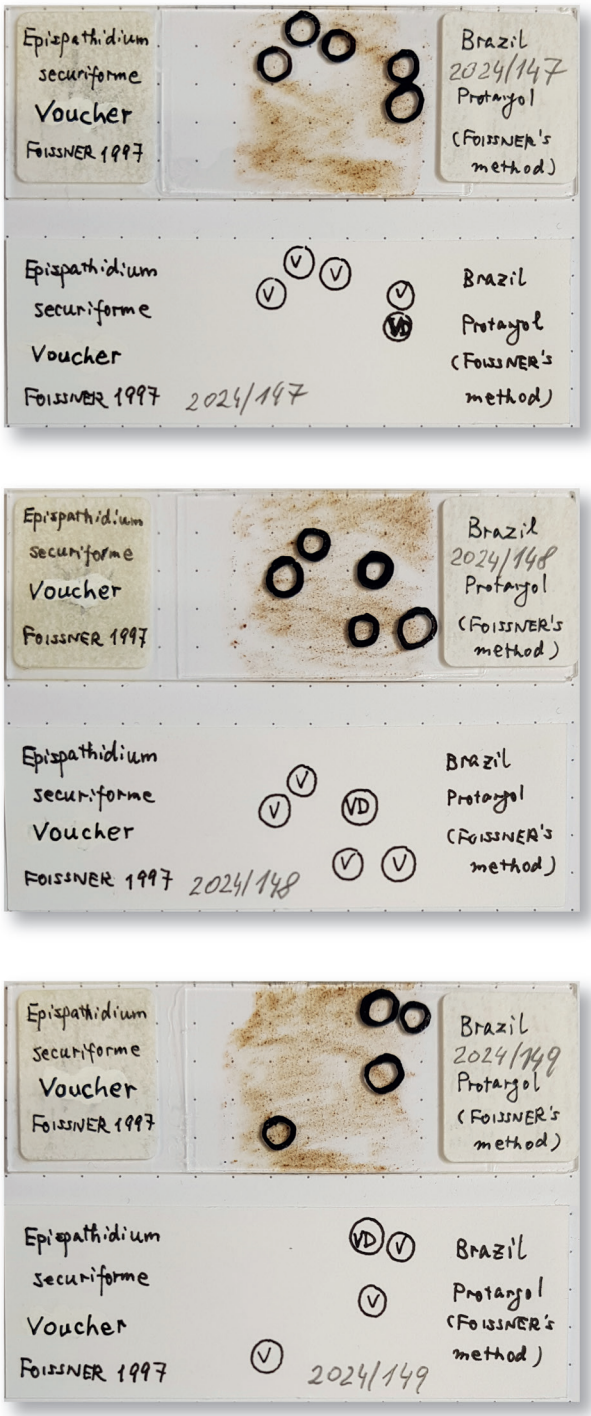
Somatic cilia about 12 µm long in vivo and rather narrowly spaced (about 2 µm; Table 6.2), especially in anterior portion of rows, where, together with the circumoral cilia, a conspicuous ciliary corona is produced; arranged in an average of 54 equidistant, bipolar, narrowly spaced kineties anteriorly forming a pronounced *Epispathidium* pattern (Fig. 6.3h, j, m, 6.4a–d, f; Table 6.2). Dorsal brush dikinetidal and three-rowed, of ordinary distinctness, that is, occupies about 31% of body length with bristles up to 5 µm long in vivo; dikinetids obliquely arranged in anterior half of brush, where distances between rows abruptly increase subapically, an unusual feature likely not caused by simple spatial constraints because the neighbouring kineties are ordinarily spaced; a fourth row comprising 11–17 dikinetids occurs ventral[13] of row 1 in two out of 32 specimens analysed. Brush rows commence with some ordinary cilia anteriorly, rows 1 and 2 end at nearly same level, continue with ordinary cilia posteriorly, and comprise an average of 55 and 50 dikinetids, respectively; brush bristles conspicuously soft, can beat up and down, anterior bristle pairs fusiform and about 5 µm long, posterior pairs with anterior bristle slightly clavate and up to 3 µm long, posterior bristle rod-shaped and about 2 µm long; row 3 shorter than rows 1 and 2 by about 78%, composed of an average of only 13 dikinetids with rod-shaped bristles, followed by a monokinetidal tail extending to second body third and composed of rather closely spaced, rod-shaped, about 2 µm long bristles (Fig. 6.3e, h, m, n, 6.4d, e; Table 6.2).

Oral bulge slanted by 45–60°, very conspicuous because on average 1.8 times longer than widest trunk region, distinctly set off from body proper and up to 6 µm high; bulge length relative to trunk width, however, highly variable (ratio 1.2–2.2:1; Table 6.2), basically, the stouter the specimens the lower the ratio and vice versa, as in Costa Rican population of *Epispathidium regium* and in an Austrian population of *Epispathidium amphoriforme* investigated recently;[14] bulge surface convex, rarely flat when cell is viewed laterally, elongate elliptical in frontal view. Circumoral kinety of same shape as oral bulge, continuous and separate from ciliary rows; composed of very narrowly spaced dikinetids each associated with a cilium, a fine fibre extending into oral bulge, and a long nematodesma. Oral basket conspicuous in protargol preparations because composed of distinct, cuneate nematodesma bundles extending to mid-body (Fig 6.3f, h–k, 6.4a–d, f; Table 6.2).

Notes on Korean population described by Jang et al. (2017): As mentioned in the Remarks section, this population was described after in vivo observations only. Body size is about 165 × 65 µm, which is distinctly smaller than the populations described by Kahl (1930a; body length 200–300 µm) and in the present work (180–250 × 50–80 µm). This difference indicates that the identification by Jang et al. (2017) is somewhat questionable.

[13] Note by H. Berger: In the raw manuscript, W. Foissner wrote that the fourth row is "right" of brush row 1. I am not certain what he exactly meant, because "to the right of" is inexact in that case. I suppose that the fourth row is ventral to row 1. "To the right of row 1" would mean that the fourth row is between row 1 and row 2. I did not check the situation in the slides.

[14] Note by H. Berger: The "recently" is somewhat misleading in the present context because W. Foissner wrote this text before about 20 years. Obviously, he meant the record of *Epispathidium amphoriforme* by Foissner et al. (2005). These data are not included in the brief review of *Epispathidium amphoriforme* (p. 197).



For further details (description and five micrographs, showing, inter alia, macronucleus and extrusomes), see Jang et al. (2017).

Resting cyst (original data from a population from the Dominican Republic): Cysts colourless, covered by an about 5 µm thick, mucilaginous layer, 41–53 µm (mean = 47.3, n = 8) in diameter; wall smooth and 1.5 µm thick; macronucleus shorter than in interphase specimens; extrusomes recognizable.

Occurrence and ecology: The type locality of *Epispathidium securiforme* is moss on stones (gneiss and slate) from the Zillertal (the northern end of this valley is about at 47.40°N 11.83°E; Tyrol, Austria), where it was common, but never abundant (Kahl 1930a, p. 380; exact sample site not mentioned). Later, he found it (with exactly the same morphology and sensitivity to coverglass pressure) in moss from an old tile roof in the Lüneburger Heide, northern Germany (Kahl 1930a).

Fig. 6.4g–l *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 (originals. Protargol slides). **g–l:** Slides (g, i, k) and protocols (h, j, l) of voucher specimens (V) and voucher specimens drawn (VD) of population from Brazil. Accession numbers (LI): 2024/147, 148, 149.

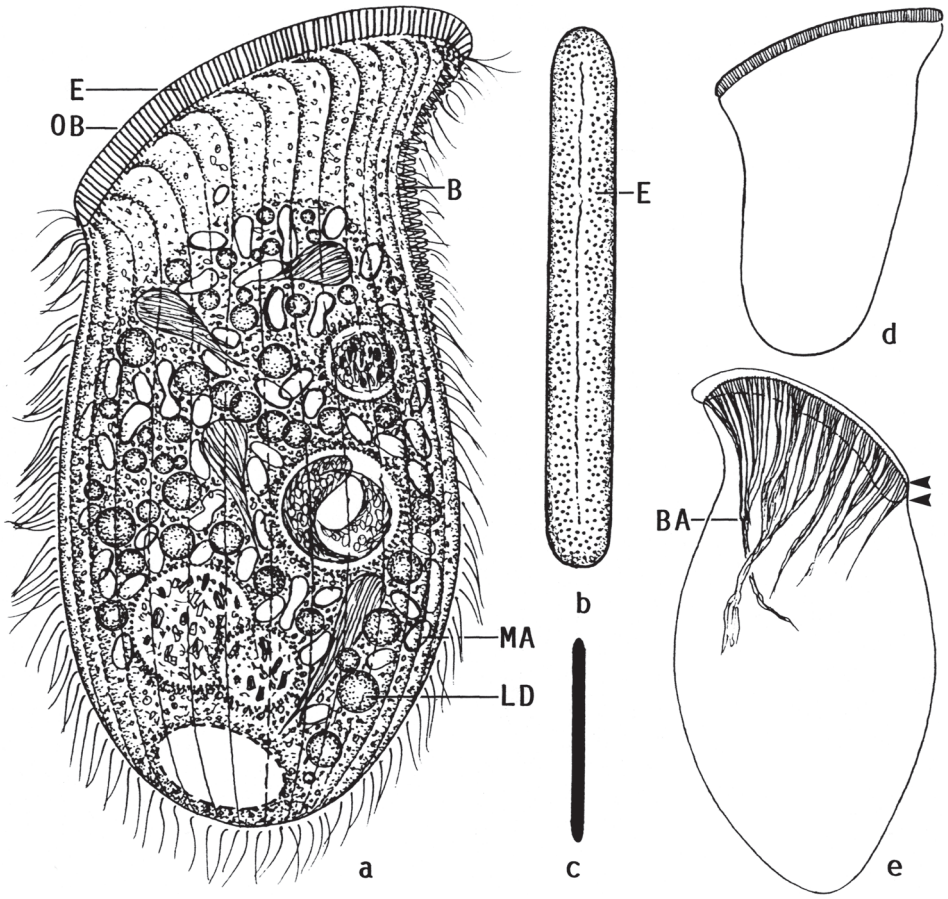
In South America, *Epispathidium securiforme* occurred with low abundance in the surroundings of Rio de Janeiro, Brazil, viz., in the shrub zone of the Restingha area about 1 km off the Atlantic Sea coast (Fig. 6.3e–n, 6.4a–f). The sample, which consisted of surface litter (0–2 cm), fine roots, and strongly decayed organic material sieved off from the upper 10 cm sand layer, was very humic, slightly saline, and had pH 5.3 (in water). In the Dominican Republic, a small population developed in a non-flooded Petri dish culture with highly saline soil and litter from a mangrove swamp.

Epispathidium salsum nov. spec.

(Fig. 6.5a–o, 6.6a–l, Table 6.3)

Nomenclature: The species-group name *sals·us*, *-a*, *-um* (Latin adjective [m, f, n]; salty, briny, saline; Brown 1954, p. 678) refers to the highly saline site the species was discovered.

Diagnosis: Body size about 130 × 60 µm in vivo. Body shape bluntly spatulate with oblique oral bulge about as long as widest trunk region. Macronucleus composed of about



110 scattered nodules. Oral bulge extrusomes rod-shaped, 5–6 µm long. On average 33 ciliary rows, 3 anteriorly modified to ordinary dorsal brush occupying about 31% of body length.

Type locality: Highly saline soil from Pacific coast at Mill Valley (37°30'N 122°30'W), San Francisco, USA.

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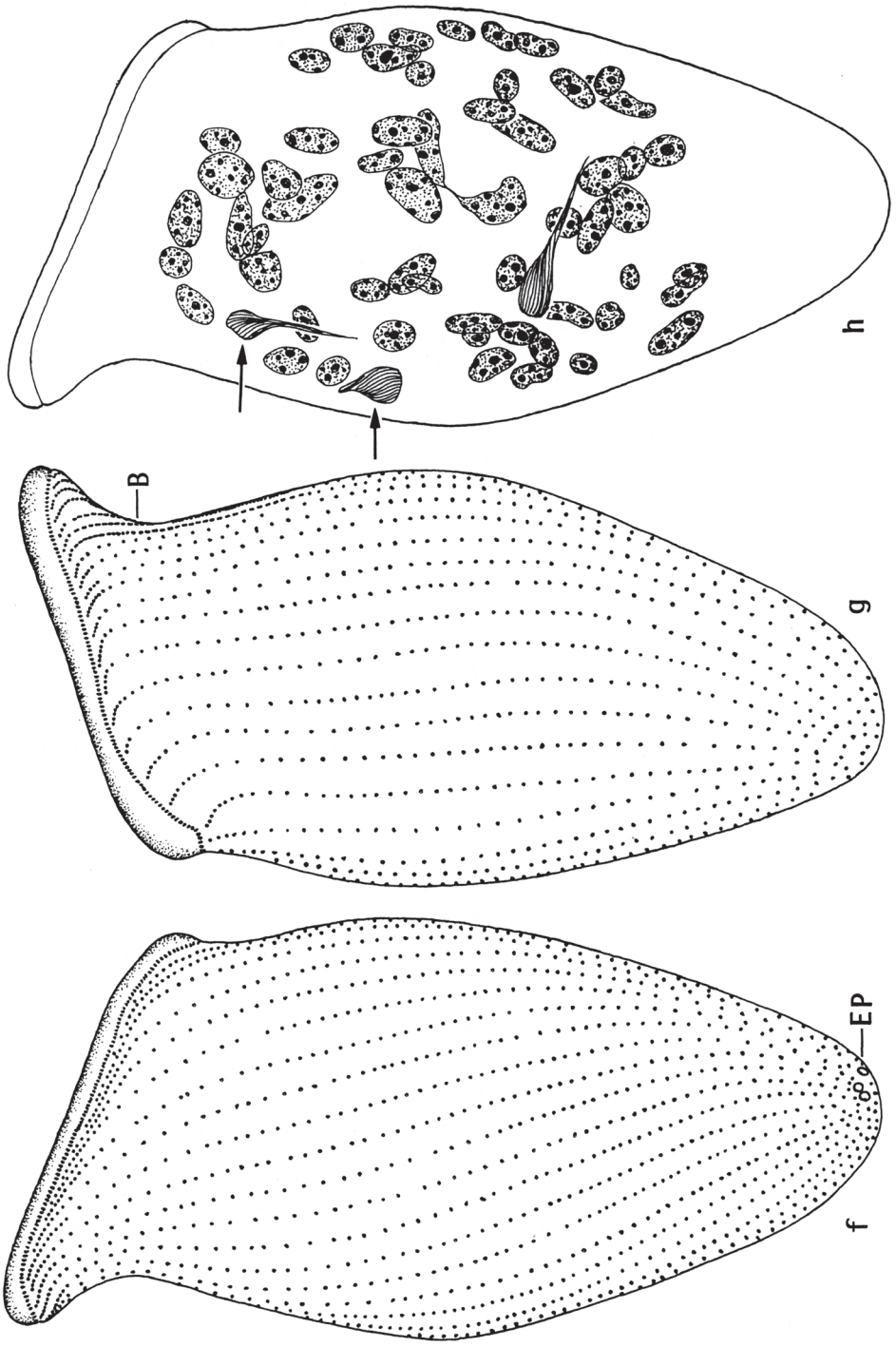
Table 6.3 Morphometric data on *Epispathidium salsum* nov. spec. (type population from USA, original data)[a]

Characteristic	Mean	M	SD	SE	CV	Min	Max	n
Body, length	109.2	103.0	13.3	3.1	12.2	97.0	145.0	19
Body, width	51.7	50.0	8.8	2.0	17.1	39.0	77.0	19
Body length:width, ratio	2.1	2.1	0.2	0.1	8.7	1.9	2.5	19
Oral bulge, length	48.3	50.0	9.8	2.2	20.2	31.0	70.0	19
Oral bulge length:body width, ratio	0.9	0.9	0.1	0.1	12.8	0.7	1.1	19
Oral bulge, height	3.5	3.5	–	–	–	3.0	4.0	19
Oral bulge, width in frontal view	7.1	7.0	1.1	0.3	15.6	6.0	10.0	15
Circumoral kinety to last dikinetid of brush row 1, distance	29.2	27.0	5.6	1.3	19.0	23.0	45.0	19
Circumoral kinety to last dikinetid of brush row 2, distance	33.5	31.0	5.8	1.3	17.2	26.0	50.0	19
Circumoral kinety to last dikinetid of brush row 3, distance	12.1	12.0	1.3	0.3	11.0	10.0	15.0	19
Macronucleus nodules, length	6.6	7.0	3.2	0.7	49.3	2.5	15.0	19
Macronucleus nodules, width	3.5	3.0	1.4	0.3	38.7	2.0	8.0	19
Macronucleus nodules, number[b]	109.8	120.0	–	–	–	50.0	156.0	19
Micronuclei, diameter	1.9	2.0	–	–	–	1.5	2.5	19
Micronuclei, number[b]	17.1	18.0	–	–	–	5.0	27.0	19
Somatic kineties, number	33.2	33.0	2.7	0.6	7.9	20.0	40.0	19
Basal bodies in a right-side kinety, number	49.2	50.0	12.7	2.9	25.8	30.0	84.0	19
Dorsal brush rows, number	3.0	3.0	0.0	0.0	0.0	3.0	3.0	19
Dikinetids in brush row 1, number	26.3	26.0	5.7	1.3	21.5	19.0	40.0	19
Dikinetids in brush row 2, number	30.1	30.0	5.5	1.3	18.2	23.0	45.0	19
Dikinetids in brush row 3, number	11.6	11.0	1.8	0.4	15.6	10.0	16.0	19

[a] Data based on mounted and protargol-prepared (Foissner's method) specimens from a non-flooded Petri dish culture. Specimens were selected for cells with ordinary nuclear apparatus (see text). Measurements in µm. CV – coefficient of variation in %, M – median, Max – maximum, Mean – arithmetic mean, Min – minimum, n – number of individuals investigated, SD – standard deviation, SE – standard error of arithmetic mean.

[b] Approximate values because number of macronuclear nodules difficult to count due to the high number and/or similar-impregnated cell inclusions.

← **Fig. 6.5a–e** *Epispathidium salsum* nov. spec. (originals. a–d, from life; e, protargol preparation). **a:** Left side view of a representative specimen, 130 µm. The cell is packed with lipid droplets, macronucleus nodules and clavate cortices of partially digested *Euglena* sp. **b:** Frontal view of oral bulge studded with extrusomes. **c:** Oral bulge extrusome, length 5–6 µm. **d:** Left side view of campanulate shape variant. **e:** Right side view showing that the oral basket extends beyond mid-body, 100 µm. Arrowheads mark circumoral kinety. B – dorsal brush, BA – oral basket, E – extrusomes, LD – lipid droplet, MA – macronucleus nodule, OB – oral bulge.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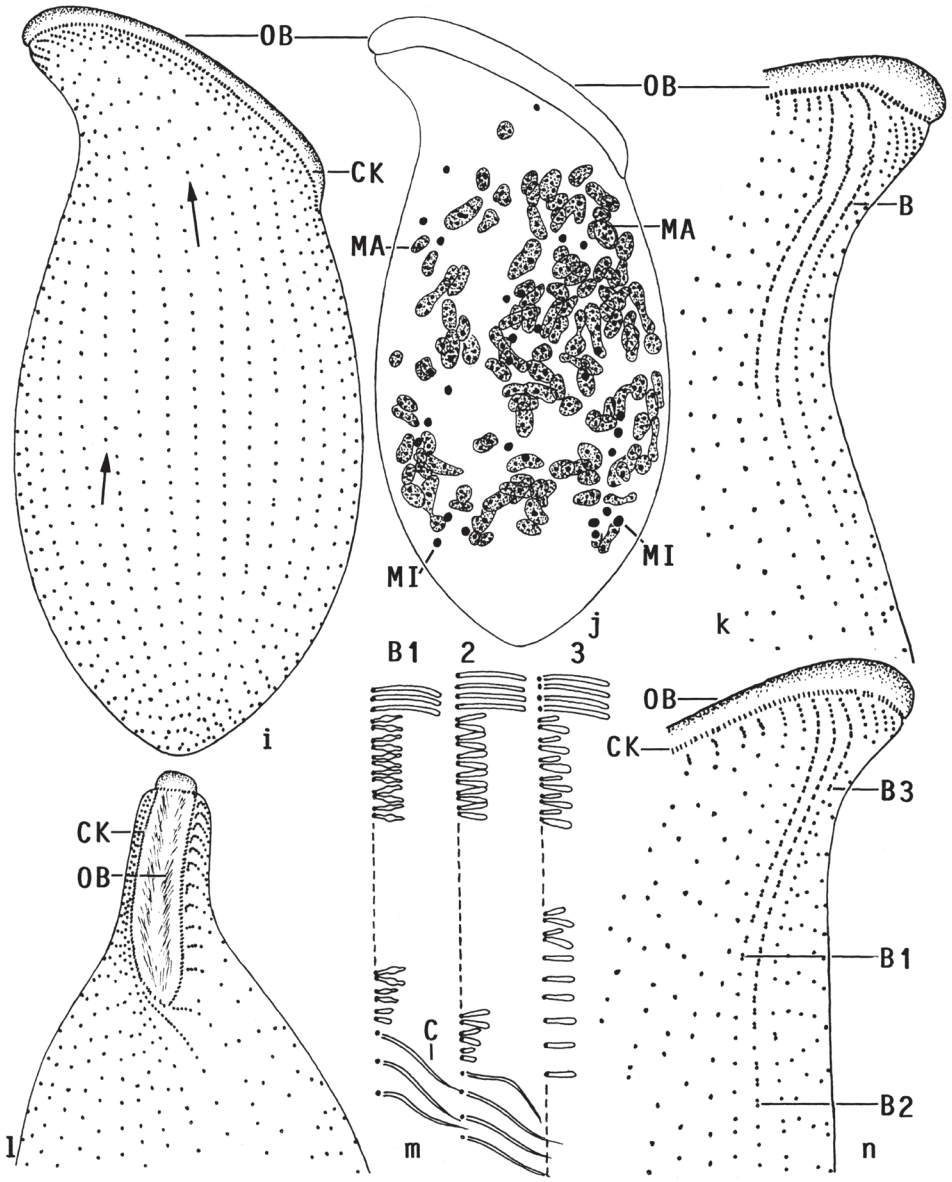

Fig. 6.5i–n *Epispathidium salsum* nov. spec. (originals. i–l, n, protargol preparation; m, from life). **i, j:** Right side ciliary pattern (i) and nuclear apparatus (j) of a specimen with meridionally extending ciliary rows, 100 µm. Arrows mark irregularities in ciliary pattern. **k, n:** Left dorsolateral views showing specimens with slightly disturbed (k) and undisturbed (n) dorsal brush, length of second brush row 33 µm and 36 µm. **l:** Ventral view showing the 30 µm long oral bulge. **m:** Anterior and posterior region of dorsal brush. B – dorsal brush, B1–3 – dorsal brush rows, C – ordinary somatic cilium, CK – circumoral kinety, MA – macronucleus nodules, MI – micronuclei, OB – oral bulge.

← **Fig. 6.5f–h** *Epispathidium salsum* nov. spec. (originals. Protargol preparation). Ciliary pattern of right (f) and left (g) side and macronucleus apparatus (h) of holotype specimen with obliquely extending right side ciliary rows, 135 µm. Arrows mark cortices of partially digested *Euglena* specimens. B – dorsal brush, EP – excretory pores.

Type material: The slide (Fig. 6.6e) containing the holotype (Fig. 6.5f–h; accession number 2024/150) and three paratype slides (Fig. 6.6g–l; accession numbers 2024/151, 152, 153) have been deposited in the Biology Centre of the Upper Austrian Museum in Linz (LI).

ZooBank registration: urn:lsid:zoobank.org:act:B98CA626-6A3E-4813-A67F-F12C5429511C

Remarks: The *Epispathidium* ciliary pattern is rather indistinct in this species because the cilia are only moderately condensed in the apical region of the kineties and few of the left side rows are curved so strongly ventrally that they run in parallel with the circumoral kinety. On the other hand, the circumoral kinety is continuous and distinctly separate from the ciliary rows in the about 40 specimens investigated. Thus, the population cannot be assigned to *Spathidium*. The taxonomic significance of the oblique arrangement of the right-side ciliary rows remains obscure. Likely, it is a population-specific feature because some specimens have ordinary, meridionally extending rows. Thus, this feature was excluded from the diagnosis.

Epispathidium salsum is rather similar to *Epispathidium regium*, differing from that species by the much less distinct *Epispathidium* ciliary pattern (see above), the body size (about 110 × 50 µm vs. 170 × 55 µm in protargol preparations), the body length:width ratio (about 2.1:1 vs. 3.1:1 in preparations), and the number of ciliary rows (33 vs. 41–48 on average), and the number of dorsal brush dikinetids (68 vs. 117 on average). *Epispathidium salsum* also resembles *Spathidium canadense* Wilbert nom. nov. (for note on new name, see *Epispathidium regium*, p. 144) which, however, has a *Spathidium* ciliary pattern, a much shorter brush, and an oral bulge about twice as long as the widest trunk region. Finally, the body

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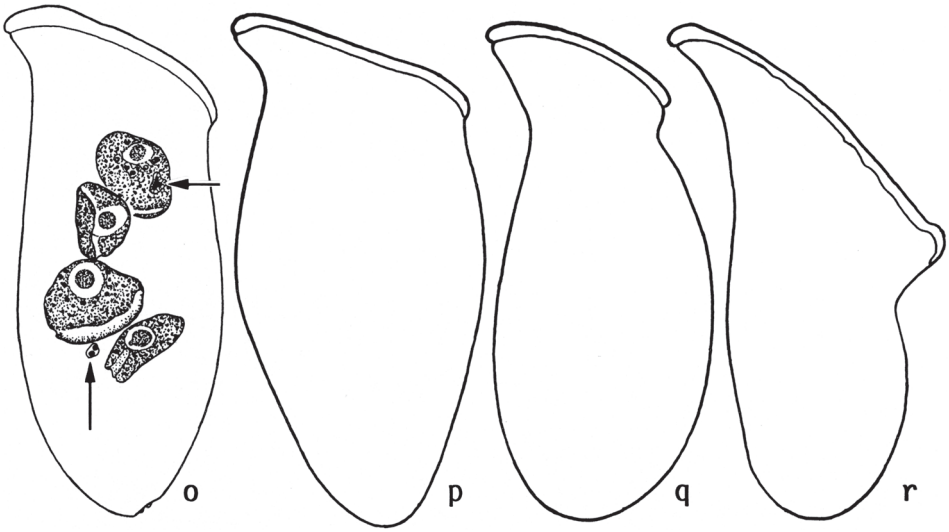


Fig. 6.5o *Epispathidium salsum* nov. spec. (original. Protargol preparation). A specimen with four large macronucleus nodules and two wrinkled micronuclei (arrows), 125 µm.
Fig. 6.5p–r Comparison of body shape of protargol-prepared, representative specimens of *Epispathidium salsum* (p), *Epispathidium regium* (q), and *Spathidium canadense* (r).

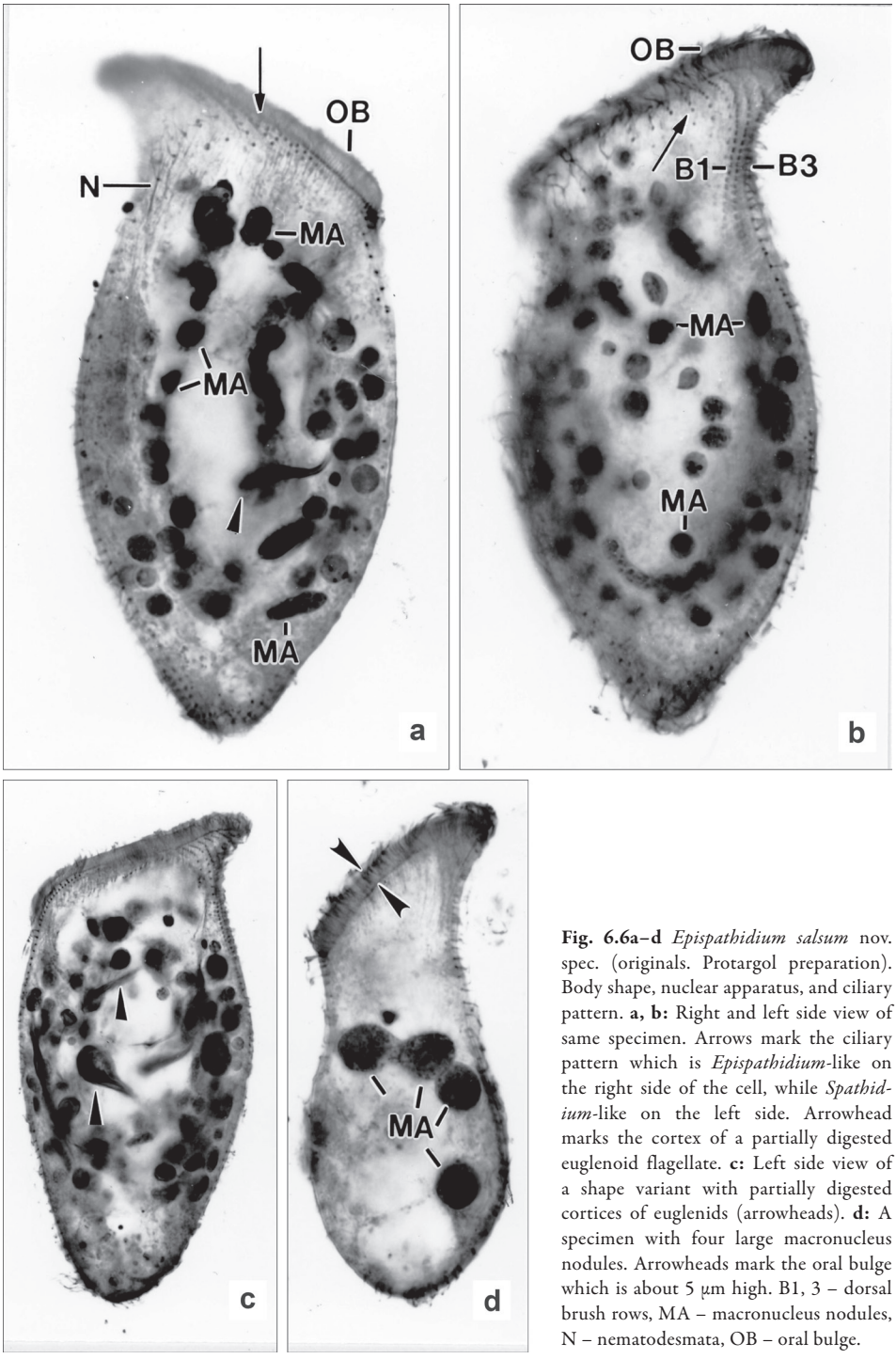


Fig. 6.6a–d *Epispathidium salsum* nov. spec. (originals. Protargol preparation). Body shape, nuclear apparatus, and ciliary pattern. **a, b:** Right and left side view of same specimen. Arrows mark the ciliary pattern which is *Epispathidium*-like on the right side of the cell, while *Spathidium*-like on the left side. Arrowhead marks the cortex of a partially digested euglenoid flagellate. **c:** Left side view of a shape variant with partially digested cortices of euglenids (arrowheads). **d:** A specimen with four large macronucleus nodules. Arrowheads mark the oral bulge which is about 5 µm high. B1, 3 – dorsal brush rows, MA – macronucleus nodules, N – nematodesmata, OB – oral bulge.

Fig. 6.6e–h *Epispathidium salsum* nov. spec. (originals. Protargol slides). **e, f:** Slide (e) and protocol (f) containing holotype (H) and paratypes (P). Accession number (LI): 2024/150. **g, h:** Slide (g) and protocol (h) containing paratypes drawn (PD) and paratypes (P). Accession number (LI): 2024/151.

shape is rather different in these three species (Fig. 6.5p–r): stout and spathidiid in *Epispathidium salsum*; of usual length:width ratio and epispathidiid in *Epispathidium regium*; and stout and axe-shaped in *Spathidium canadense*. Nonetheless, care is needed in identifying these species because they are rather similar in size and shape of body and extrusomes and, especially, the nuclear pattern.

Description: Body size 100–160 × 45–90 µm in vivo, usually about 130 × 60 µm; length:width ratio 1.9–2.5:1, on average 2.1:1 in protargol preparations (Table 6.3). Body shape bluntly spatulate, rarely amphoriform or campanulate with slightly to ordinarily oblique oral bulge, short neck, and narrowly to broadly rounded posterior body end; widest usually in mid-body much more narrowing posteriorly than anteriorly; laterally flattened up to 2:1 (Fig. 6.5a, d, e, h, i, 6.6a–d). Nuclear apparatus scattered throughout body, except for apical and terminal regions. Usually about 110 macronucleus nodules of very different shape and size, as shown in Fig. 6.5h, j. Micronuclei about 3 µm across in vivo, spherical to broadly ellipsoidal, usually impregnate deeply with protargol. About 40% of specimens have only four large, discoidal, rather faintly impregnated macronucleus nodules with a conspicuous nucleolus each; micronuclei are lacking or resorbed to some wrinkled structures surrounded by a distinct membrane (Fig. 6.5o). These specimens, which have an ordinary ciliary pattern, are smaller on average (minimum body length 60 µm) and contain few or no food inclusions. Contractile vacuole in posterior body end, some excretory pores in pole area. Only one type of extrusomes studded in oral bulge and scattered in cytoplasm, do not impregnate with the protargol method used, not even those in the cytoplasm.

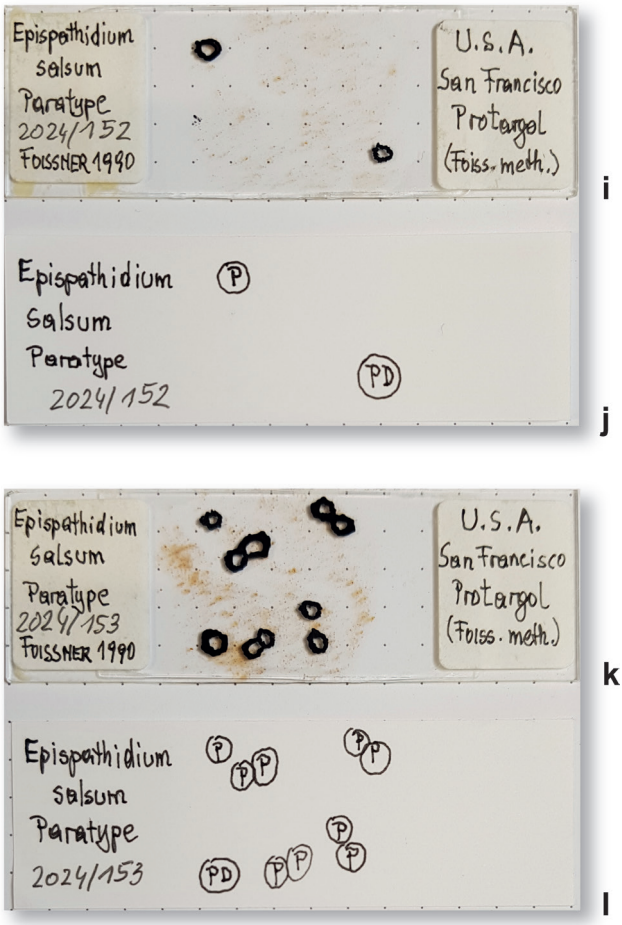


Fig. 6.6i–l *Epispathidium salsum* nov. spec. (originals. Protargol slides). **i–l:** Slides (i, k) and protocols (j, l) containing paratypes drawn (PD) and paratypes (P). Accession numbers (LI): 2024/152, 153.

Individual extrusomes inconspicuous, that is, rod-shaped with bluntly pointed ends and 5.0–6.0 × 0.3–0.4 µm in size (Fig. 6.5a–c). Cortex very flexible, contains about eight rows of colourless granules between each two kineties; individual granules compact and thus rather refractive in vivo, about 0.8 × 0.4 µm in size. Cytoplasm colourless, contains some food vacuoles and many lipid droplets up to 10 µm across in well-nourished specimens. Feeds on hypotrichous ciliates digested in up to 60 µm-sized vacuoles and on *Euglena* sp., whose cortex becomes conspicuously drumstick shaped during digestion (Fig. 6.5a, h). Glides and swims moderately rapidly on microscope slide.

Somatic cilia about 10 µm long in vivo, arranged in an average of 33 bipolar, ordinarily spaced and ciliated rows extending meridionally on left side, while more or less obliquely on right in most specimens (Fig. 6.5f, g); rarely, rows extend meridionally on both sides and have small irregularities, for instance, overlapping breaks (Fig. 6.5i). Ciliary rows distinctly separate from circumoral kinety in 40 specimens investigated, *Epispathidium* pattern, however, rather indistinct on left side because cilia only slightly condensed apically, and most rows fairly inconspicuously curved ventrally. Right side ciliary rows densely ciliated subapically, while loosely so apically and in neck area, those near ventral side strongly curved dorsally and extending side by side with circumoral kinety, producing distinct *Epispathidium* pattern (Fig. 6.5a, f, g, i, 6.5a, b; Table 6.3). Brush on dorsal side, of ordinary structure and distinctness, that is, bristles up to 3 µm long and longest row 2 occupying 31% of body length on average; frequently with small irregularities, such as minute and/or overlapping breaks and some dikinetids out of line (Fig. 6.5a, g, k, m, n, 6.6b; Table 6.3). Rows 1 and 2

of nearly same length, row 3 shortened by almost two thirds, but with monokinetidal tail of 3 µm long bristles extending behind mid-body; anterior portion of rows not composed of ordinary cilia, as usual, but of 3–7, about 7 µm long, rod-shaped bristles. Bristles of individual dikinetids of similar length in rows 1 and 2, those of row 1 fusiform, those of row 2 slightly inflated distally; anterior bristle of row 3 dikinetids slightly shorter than the 3 µm long, distally inflated posterior bristle.

Oral bulge slanted by only 15–35° and usually slightly sigmoidal, in frontal view oblong with both ends rounded; conspicuous both in vivo and in preparations because about as long as widest trunk region, circa 5 µm high, and bright due to the many refractive extrusomes contained (Fig. 6.5a, d–f, i, 6.6a; Table 6.3). Circumoral kinety oblong with bluntly pointed ventral end, distinctly separate from ciliary rows, usually except for one or few kineties in variable position; composed of narrowly spaced dikinetids forming a continuous, conspicuous row. Individual dikinetids associated with a 12 µm long cilium and a rather fine, wrinkled (by preparation?) nematodesma extending beyond mid-body in dorsal half of cell; oral basket thus conspicuous, but often only faintly impregnated (Fig. 6.5e–g, i, l, n, 6.6a; Table 6.3).

Occurrence and ecology: As yet found only at the type locality (see above), that is, a highly saline (>20‰) soil sample with pH 6.5 from the USA. *Epispathidium salsum* was moderately abundant in the non-flooded Petri dish culture. As the sample site is flooded from time to time, we cannot decide whether *Epispathidium salsum* is a marine or terrestrial species.

Epispathidium papilliferum (Kahl, 1930) Foissner, 1984

(Fig. 6.7a–k, 6.8a–v, 6.9a–l, 6.10a–q, 6.11a–o, 6.12a–p, Tables 6.4, 6.5)

1930 ***Spathidium papilliferum* spec. n.** – Kahl, Arch. Protistenk. 70: 386, Fig. 7f, f_1, 9l (Fig. 6.7g–i; original description; no type material available, see nomenclature).

1930 ***Spathidium papilliferum* Kahl, 1930** – Kahl, Tierwelt Dtl. 18: 164, Fig. 24_7 (redrawing of Fig. 6.7g; revision of ciliates).

1943 ***Spathidium papilliferum* Kahl** – Kahl, Infusorien, p. 26, Tafel VI, Fig. 3 (redrawing of Fig. 6.7g; revision of ciliates).

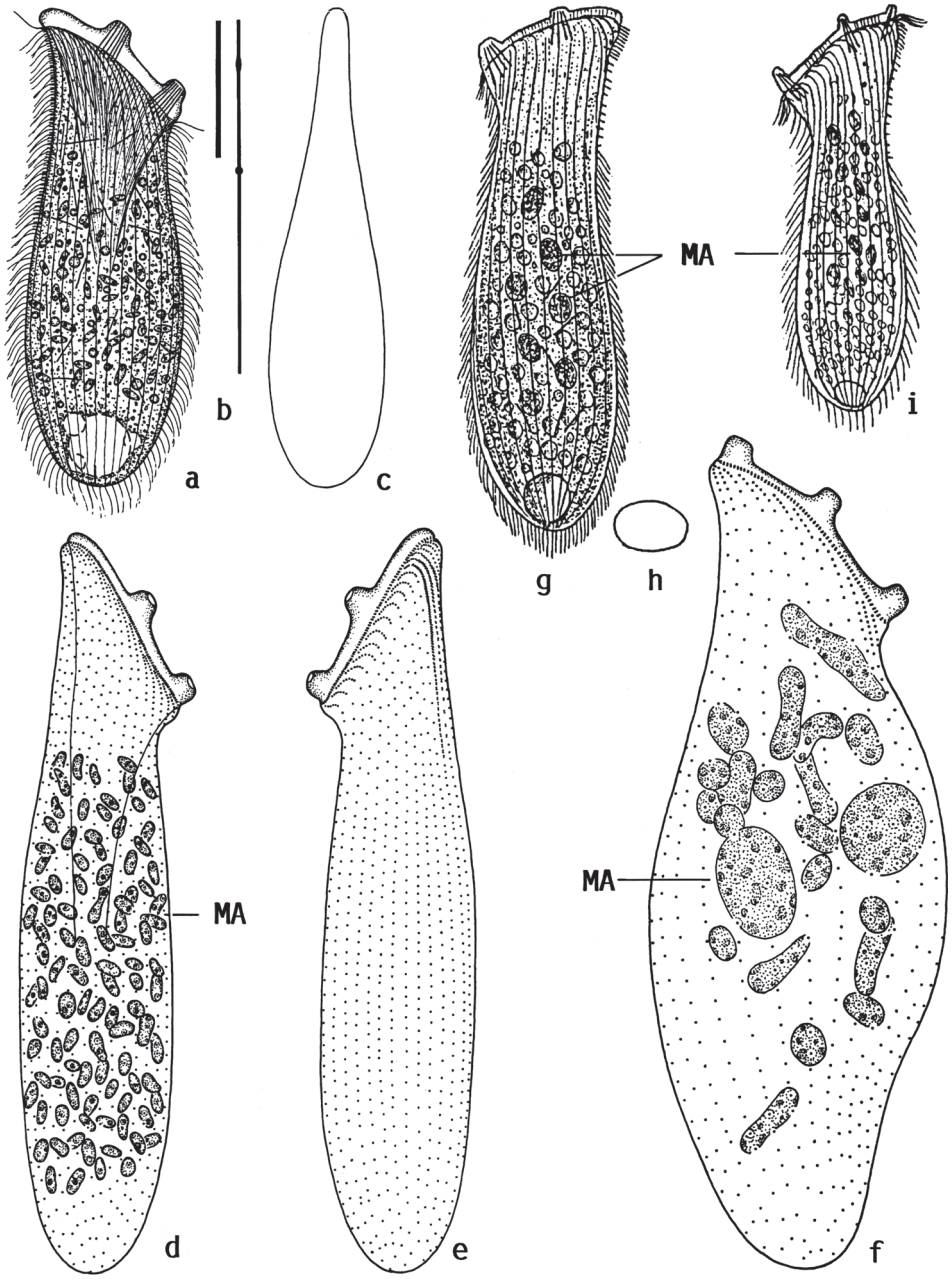
1984 ***Epispathidium papilliferum* (Kahl, 1930) nov. comb.** – Foissner, Stapfia 12: 84, Abb. 43a–h, Tabelle 21 (Fig. 6.7a–f, j, k; description of German population and Austrian population; combination with *Epispathidium*; a voucher slide with protargol-prepared specimens from Germany [accession number 1984/65; Aescht 2008, p. 171] is deposited in the Upper Austrian Museum in Linz [LI], see nomenclature).

2007 ***Vartospathidium papilliferum* comb. n.** – Jankowski, Phylum Ciliophora, p. 565 (fixation as type species of *Vartospathidium* Jankowski, 2007; combination with *Vartospathidium*).

Fig. 6.7a–i *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (a–f, from Foissner 1984; g–i, from Kahl 1930a. a–c, g–i, from life; d–f, protargol preparation. a–e, population from Germany; f, specimen from Austria; g, h, population from Mittenwald, Germany; i, type population, from Zillertal, Austria). **a:** Right side view of a representative specimen, 150 µm. **b:** Resting (10–12 × 0.5 µm; left figure) and exploded (length 30 µm; right figure) extrusomes. Shape of extrusomes not studied in detail, according to the original notes; thus, they might have been slightly acicular, as shown, for instance, in Fig. 6.9e. **c:** Dorsal view. **d, e:** Right (d) and left (e) side view of ciliary pattern, 129 µm. The macronucleus consists of many nodules. **f:** Right side view of ciliary pattern of a specimen with three papillae, 105 µm. **g–i:** Left side (g, i) and transverse (h) views, 170–260 µm. MA – macronucleus nodules. →

2017 ***Spathidium papilliferum* Kahl, 1930** – Jang, Vďačný, Shazib & Shin, J. nat. Hist. 51: 951, Fig. 5a–k, 6a–j, 7a–i, Table 2 (description of Korean population from life, see nomenclature and remarks).

Nomenclature: No derivation of the name is given in the original description or later works. The species-group name *papillifer·us*, *-a*, *-um* (Latin adjective [m, f, n]; bearing papillae) is a



composite of the Latin noun *papilla* (wart; Hentschel & Wagner 1996, p. 455), the thematic vowel *-i-* (Werner 1972, p. 37), and the Latin verb *ferre* (carry, bear; Brown 1954, p. 187); it obviously refers to the unique feature of this species, viz., the papilla-like extrusome protuberances (warts) on the oral bulge.

Kahl (1930a) did not make permanent preparations and therefore no type material is available. Foissner (1984, p. 8) obviously deposited one protargol slide (accession number 1984/65) in the Biology Centre of the Upper Austrian Museum in Linz (see list above). However, the neotypification by Foissner (1984) mentioned in Aescht (2008, p. 171) is invalid (for details on a valid neotypification, see ICZN 1999, Article 75; see also Aescht 2008, p. 132). We do not neotypify *Epispathidium papilliferum* because (i) its identity is not threatened; (ii) Foissner's (1984) redescription and preparations are of a quality allowing the specific features to be clearly recognized; (iii) Foissner (1984) deposited reference

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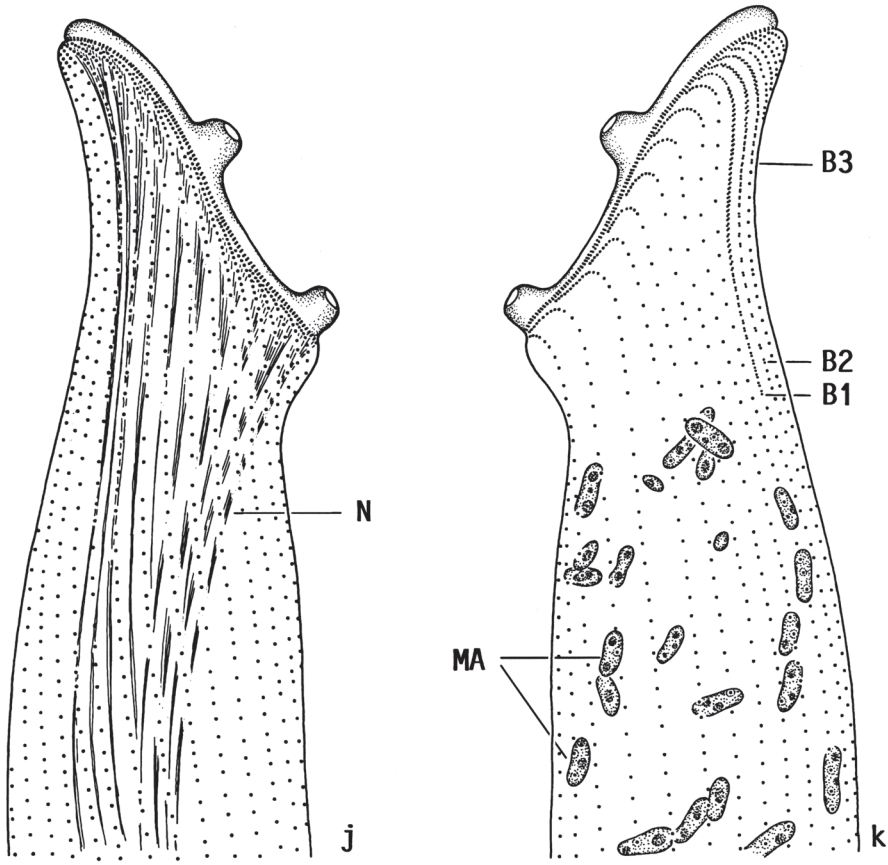

Fig. 6.7j, k *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (from Foissner 1984. Protargol preparation). Right (j) and left (k) side view of ciliary pattern in anterior body portion of a German specimen with two oral papillae and many scattered macronucleus nodules. B1–3 – dorsal brush rows, MA – macronucleus nodules, N – nematodesmata.

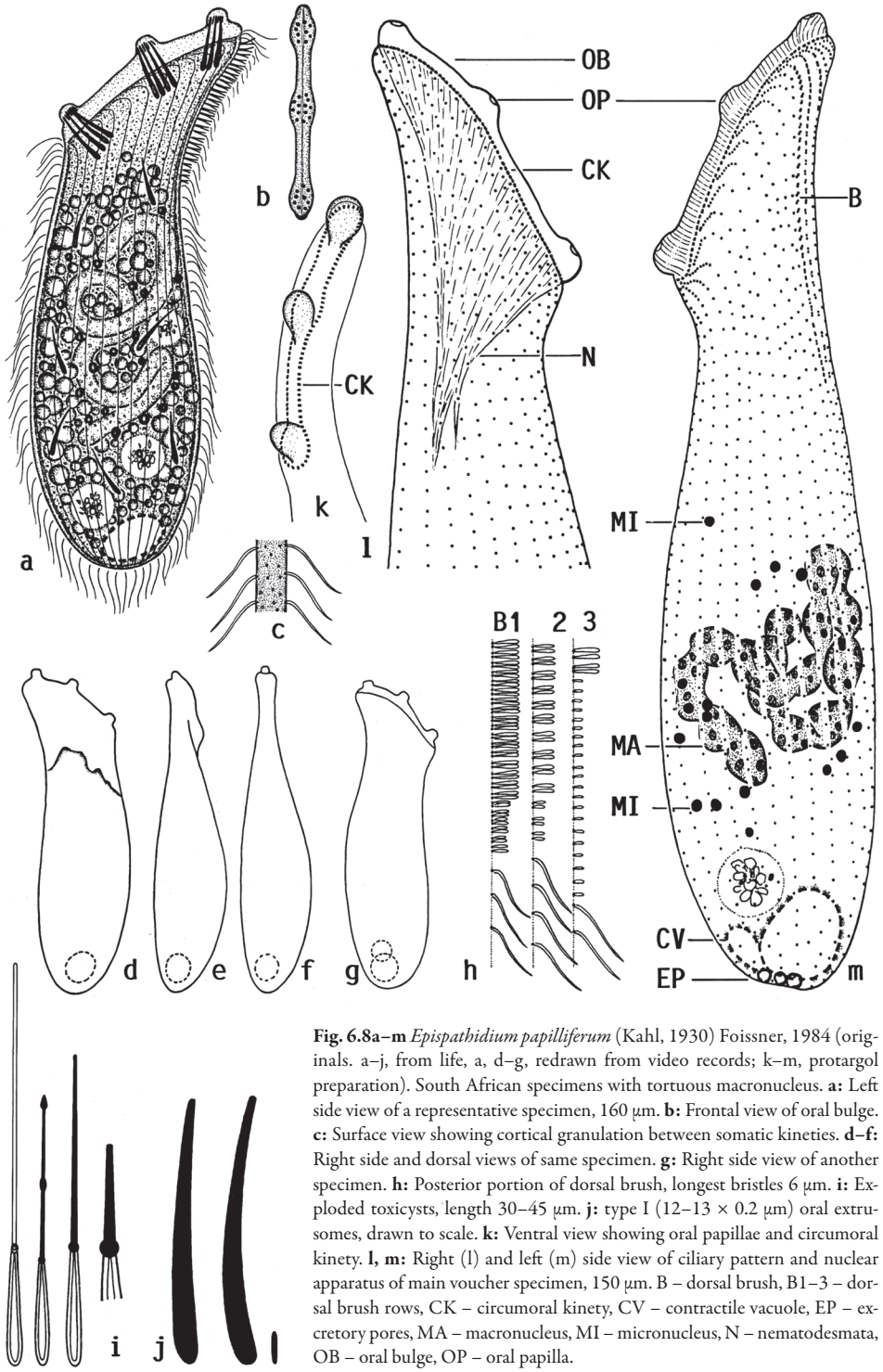

Fig. 6.8a–m *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. a–j, from life, a, d–g, redrawn from video records; k–m, protargol preparation). South African specimens with tortuous macronucleus. **a:** Left side view of a representative specimen, 160 µm. **b:** Frontal view of oral bulge. **c:** Surface view showing cortical granulation between somatic kineties. **d–f:** Right side and dorsal views of same specimen. **g:** Right side view of another specimen. **h:** Posterior portion of dorsal brush, longest bristles 6 µm. **i:** Exploded toxicysts, length 30–45 µm. **j:** type I (12–13 × 0.2 µm) oral extrusomes, drawn to scale. **k:** Ventral view showing oral papillae and circumoral kinety. **l, m:** Right (l) and left (m) side view of ciliary pattern and nuclear apparatus of main voucher specimen, 150 µm. B – dorsal brush, B1–3 – dorsal brush rows, CK – circumoral kinety, CV – contractile vacuole, EP – excretory pores, MA – macronucleus, MI – micronucleus, N – nematodesmata, OB – oral bulge, OP – oral papilla.

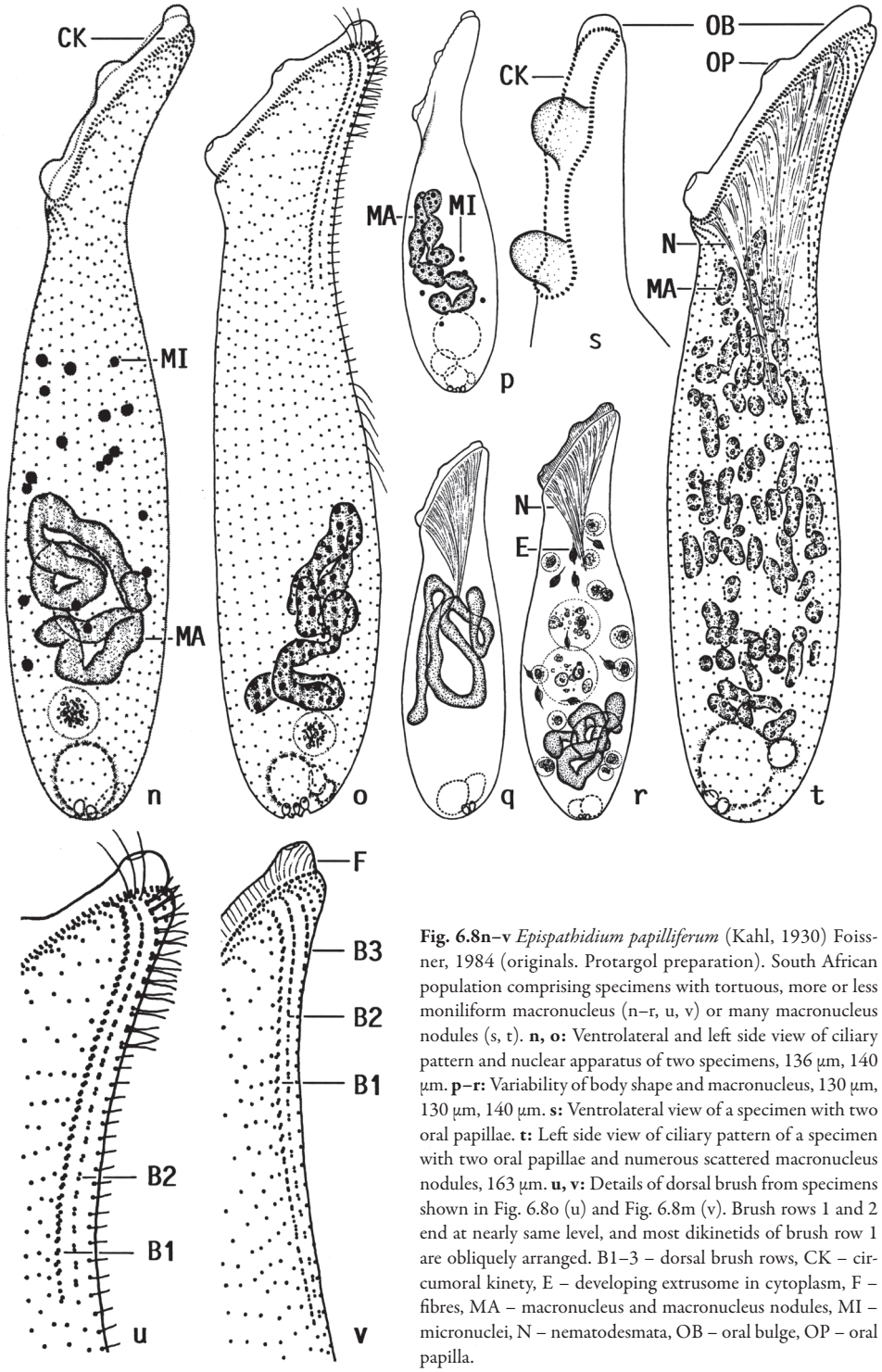

Fig. 6.8n–v *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. Protargol preparation). South African population comprising specimens with tortuous, more or less moniliform macronucleus (n–r, u, v) or many macronucleus nodules (s, t). **n, o:** Ventrolateral and left side view of ciliary pattern and nuclear apparatus of two specimens, 136 µm, 140 µm. **p–r:** Variability of body shape and macronucleus, 130 µm, 130 µm, 140 µm. **s:** Ventrolateral view of a specimen with two oral papillae. **t:** Left side view of ciliary pattern of a specimen with two oral papillae and numerous scattered macronucleus nodules, 163 µm. **u, v:** Details of dorsal brush from specimens shown in Fig. 6.8o (u) and Fig. 6.8m (v). Brush rows 1 and 2 end at nearly same level, and most dikinetids of brush row 1 are obliquely arranged. B1–3 – dorsal brush rows, CK – circumoral kinety, E – developing extrusome in cytoplasm, F – fibres, MA – macronucleus and macronucleus nodules, MI – micronuclei, N – nematodesmata, OB – oral bulge, OP – oral papilla.

material in the Biology Centre of the Upper Austrian Museum in Linz, and (iv) our new data show that *Epispathidium papilliferum* is perhaps a cluster of two or more (sub)species. We deposit voucher slides with protargol-prepared specimens from the South African population (four voucher slides, Fig. 6.12a–h; accession numbers 2024/154, 155, 156, 157) and North American (USA) population (four voucher slides, Fig. 6.12i–p; accession numbers 2024/158, 159, 160, 161) in the collection mentioned above. One of us (H. Berger) checked the whereabout of the voucher slide(s) of the population from Seekirchen, Salzburg, Austria

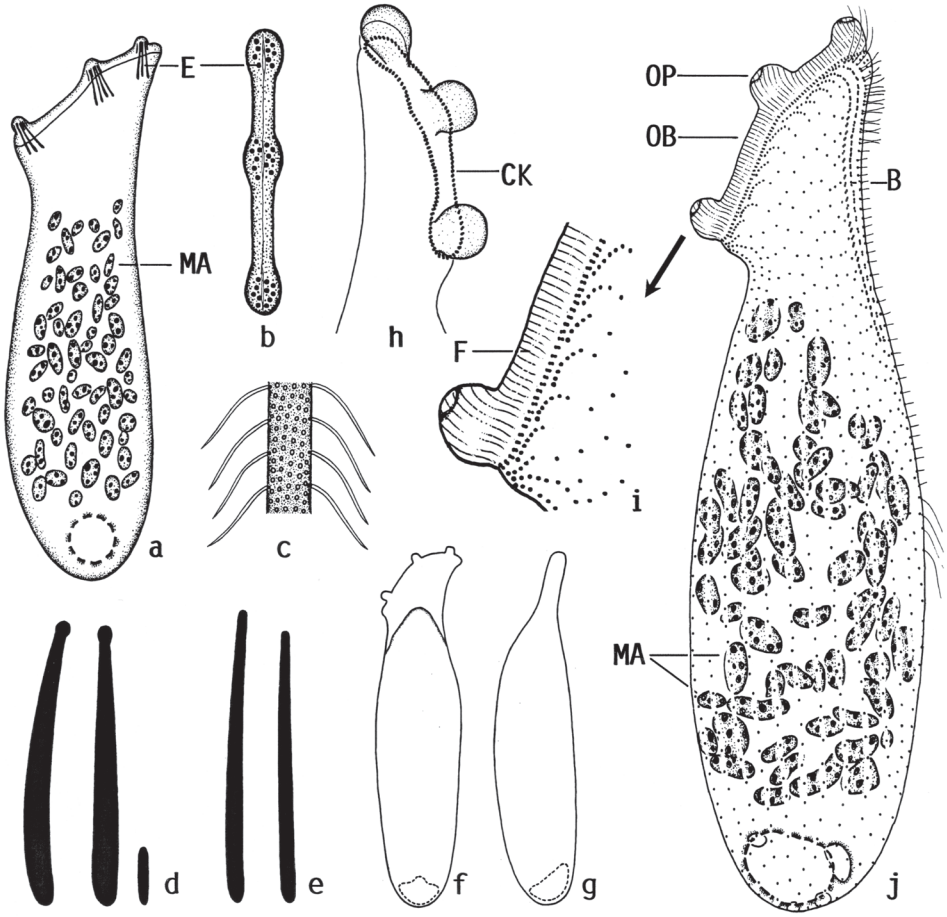

Fig. 6.9a–j *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. a–d, f–j, population from USA; e, population from Austria. a–g, from life [f, g, redrawn from video records]; h–j, protargol preparation). **a:** Outline in left side view of a representative specimen, 150 µm. **b:** Frontal view of oral bulge containing extrusomes only in the papillae. **c:** Cortical granulation between ciliary rows. **d:** Acicular type I (10.0 × 1.0 µm) and rod-shaped type II (length 2 µm) oral bulge extrusomes of a specimen from the USA, drawn to scale. **e:** The Austrian specimens have only type I extrusomes, which are indistinctly acicular and about 12.0 × 0.7 µm in size. **f, g:** Left (f) side and dorsal (g) view of a specimen with distinctly curved oral bulge. **h:** Ventrolateral view showing oral papillae and circumoral kinety. **i, j:** Left side view of ciliary pattern and nuclear apparatus, 138 µm. B – dorsal brush, CK – circumoral kinety, E – extrusomes, F – fibres, MA – macronucleus nodules, OB – oral bulge, OP – oral papilla.

(Fig. 6.7f; see Foissner 1984, p. 84). However, we could not locate them within a reasonable time, neither in the Biology Centre of the Upper Austrian Museum in Linz (LI) nor in the private archive of W. Foissner in Salzburg.

Note that Jang et al. (2017, p. 951, 955) did not mention the original description by Kahl (1930a), but the revision of Kahl (1930b). In addition, the major part of the description by Foissner (1984) is not based on an Austrian population, as indicated by Jang et al. (2017), but on a population from Germany. Further, the Chinese workers obviously overlooked the transfer of *Spathidium papilliferum* to *Vartospathidium* by Jankowski (2007).

Diagnosis (based on literature data and new observations on four populations): Body size usually about 140–170 × 30–50 µm in vivo. Body shape spatulate with oblique oral bulge about as long as widest trunk region and containing two or three extrusome

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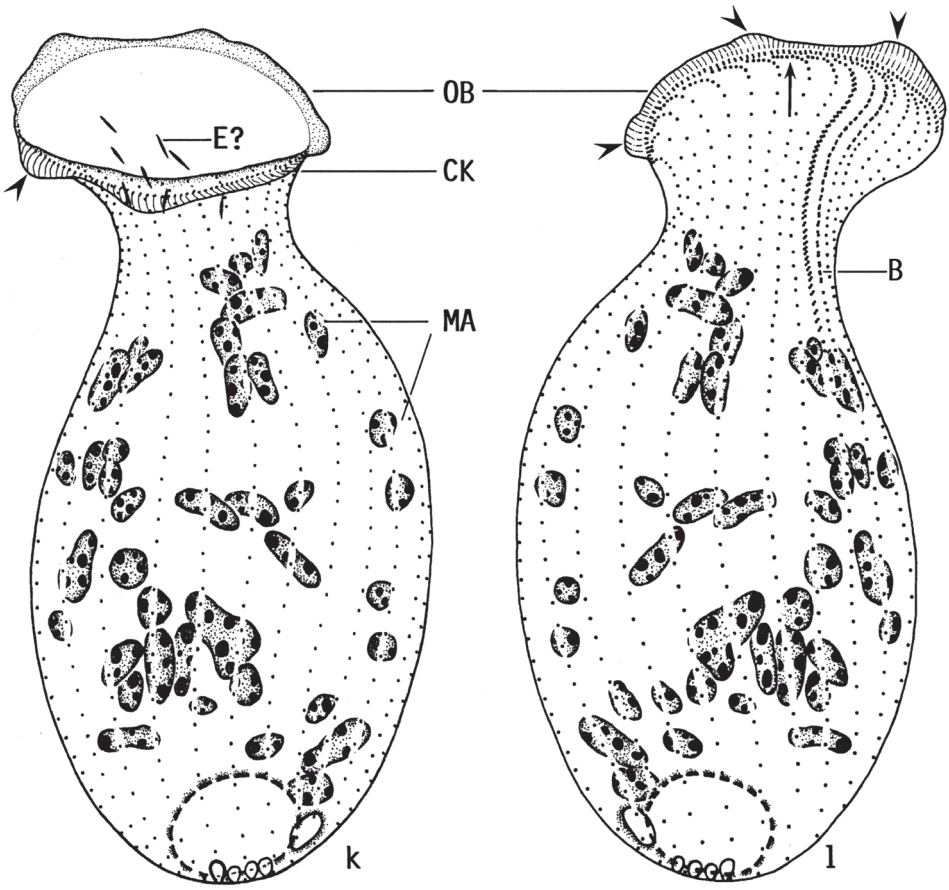

Fig. 6.9k, l *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of population from the USA. Protargol preparation). Specimen with widely open oral bulge (mouth), likely enclosing a large prey item lost by the preparation procedures, 100 µm. Arrowheads mark split oral papilla. Arrow in (l) denotes the *Epispathidium* ciliary pattern, which is maintained even if the oral bulge is opened widely. B – dorsal brush, CK – circumoral kinety, E? – extrusomes(?), MA –macronucleus nodules, OB – oral bulge.

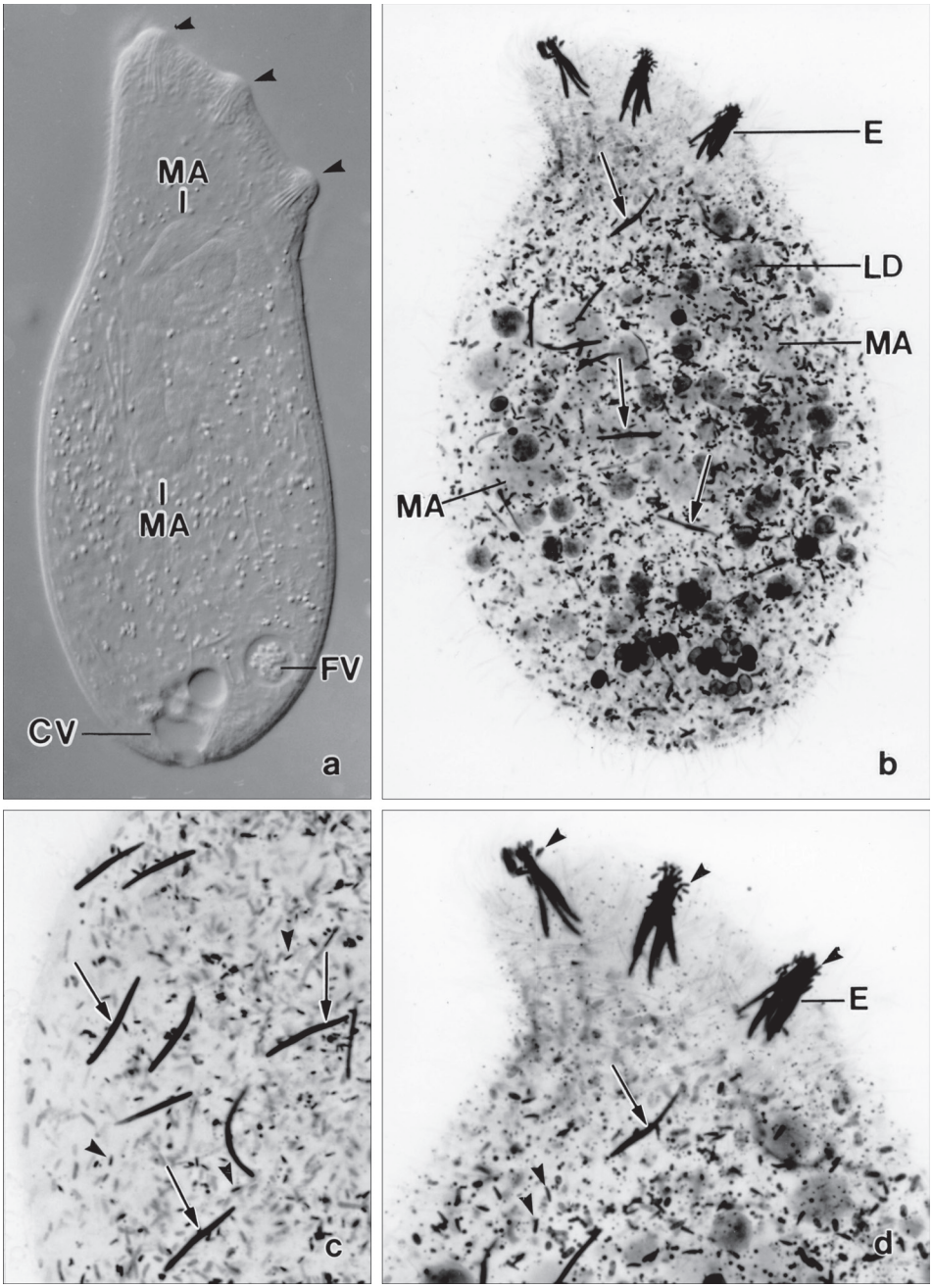

Fig. 6.10a–d *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of population from South Africa. a, from life; b–d, silver carbonate preparation). **a:** Right side view of a slightly squeezed specimen showing the three extrusome papillae (arrowheads) and the tortuous macronucleus. **b–d:** Long (arrows) and short (arrowheads) extrusomes in the oral papillae and cytoplasm. CV – contractile vacuole, E – extrusomes, FV – food vacuole, LD – lipid droplets, MA – macronucleus.

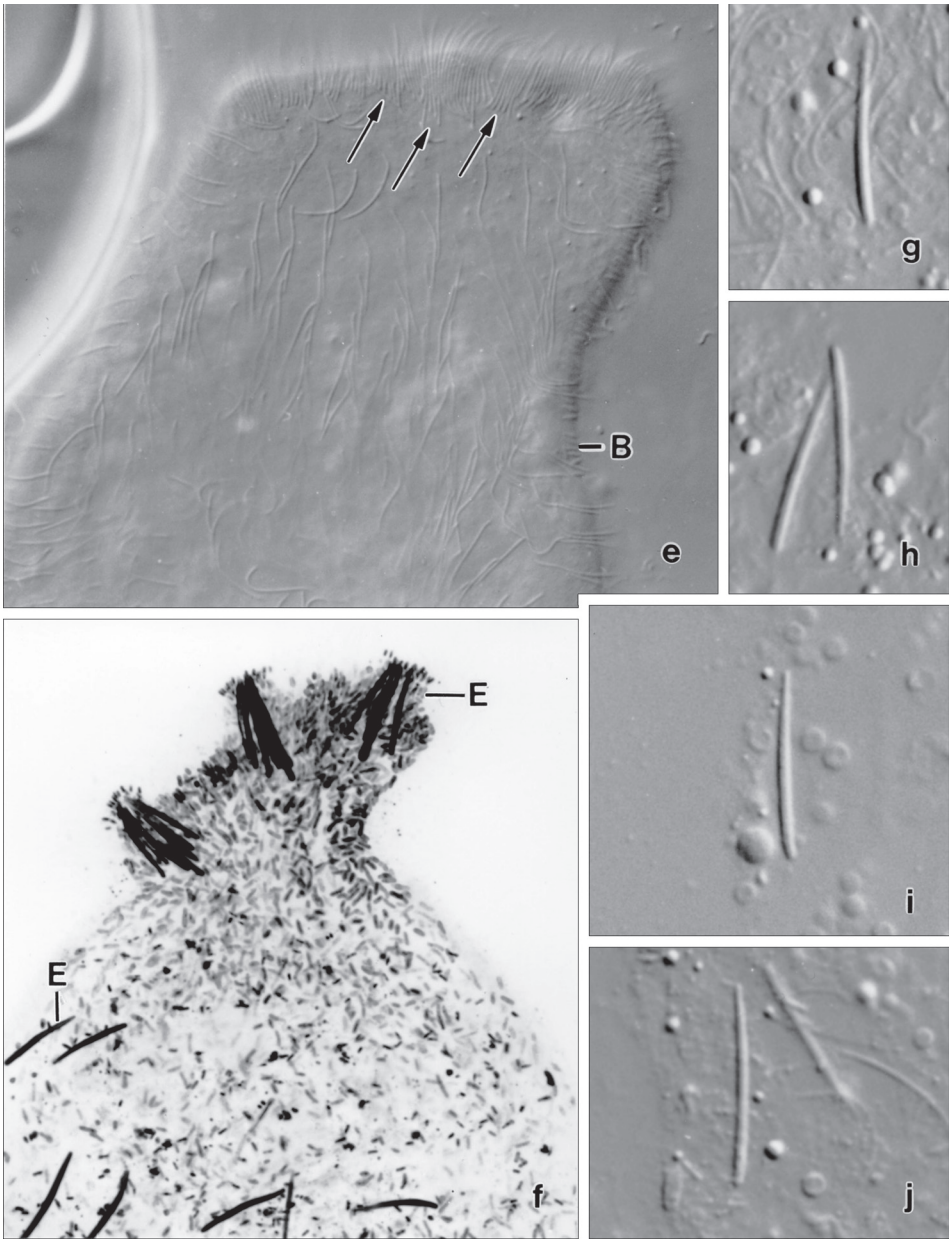

Fig. 6.10e–j *Episathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of population from South Africa. e, g–j, from life; f, silver carbonate preparation). **e:** Left side view of a squeezed specimen showing the *Episathidium* ciliary pattern (arrows) and the dorsal brush. **f:** Left side view of the anterior body portion showing long and short extrusomes in the oral papillae and in the cytoplasm. **g–j:** The long extrusomes are acicular, slightly curved, and about 12–13 × 1 µm in size. B – dorsal brush, E – extrusomes.

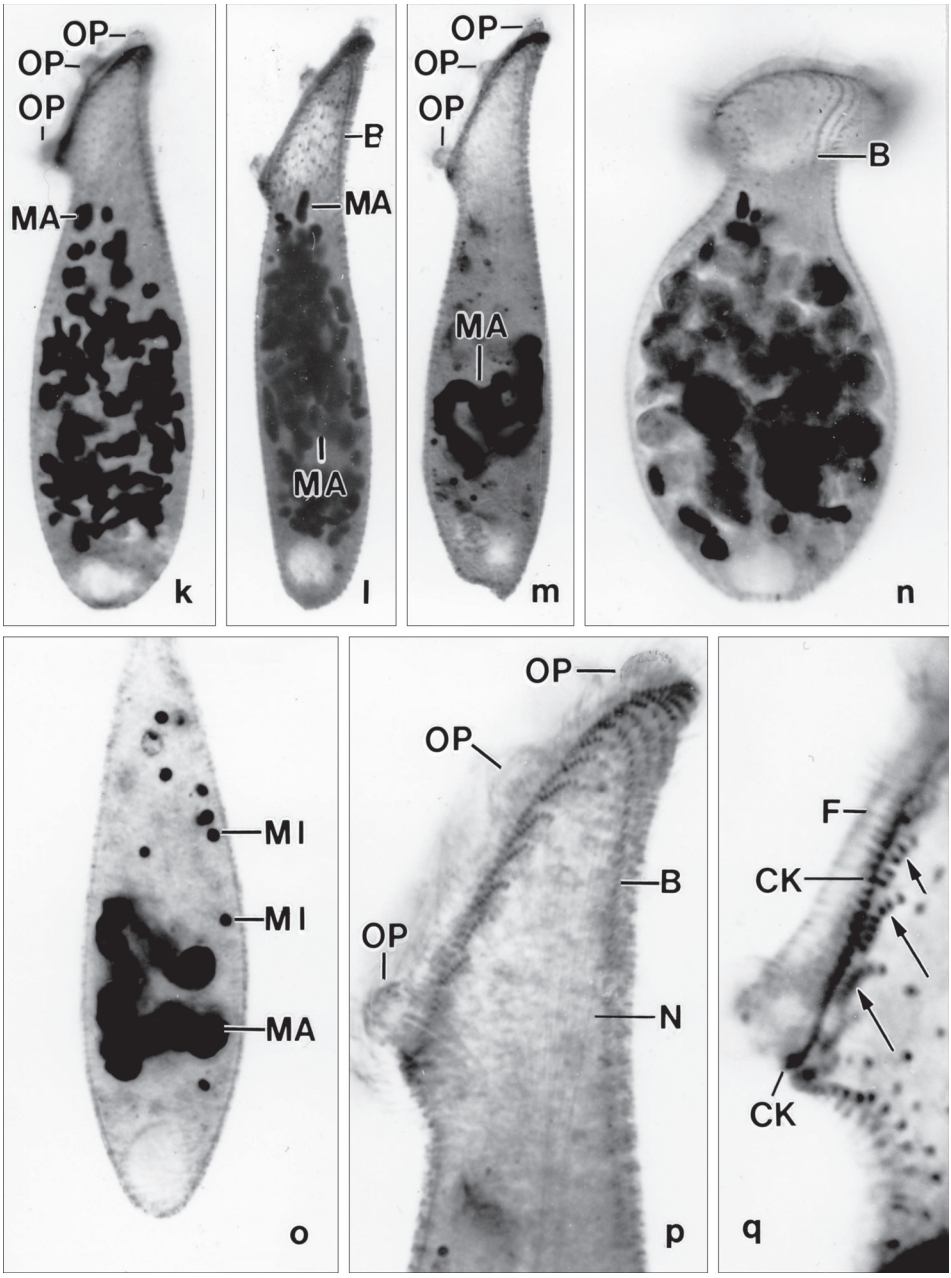

Fig. 6.10k–q *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (k, n, q, originals of population from USA; l, m, o, p, originals of population from South African. Protargol preparation). **k, q:** Same specimen with three oral papillae and many macronucleus nodules. Note the pronounced *Epispathidium* ciliary pattern (arrows in q). **l:** A specimen with two oral papillae and many macronucleus nodules. **m, p:** Same specimen with three oral papillae and a tortuous macronucleus. **n:** A specimen with widely opened mouth showing the *Epispathidium* ciliary pattern maintained. **o:** Nuclear apparatus. B – dorsal brush, CK – circumoral kinety, F – fibres, MA – macronucleus and macronucleus nodules, MI – micronuclei, N – nematodesmata, OP – oral papillae.

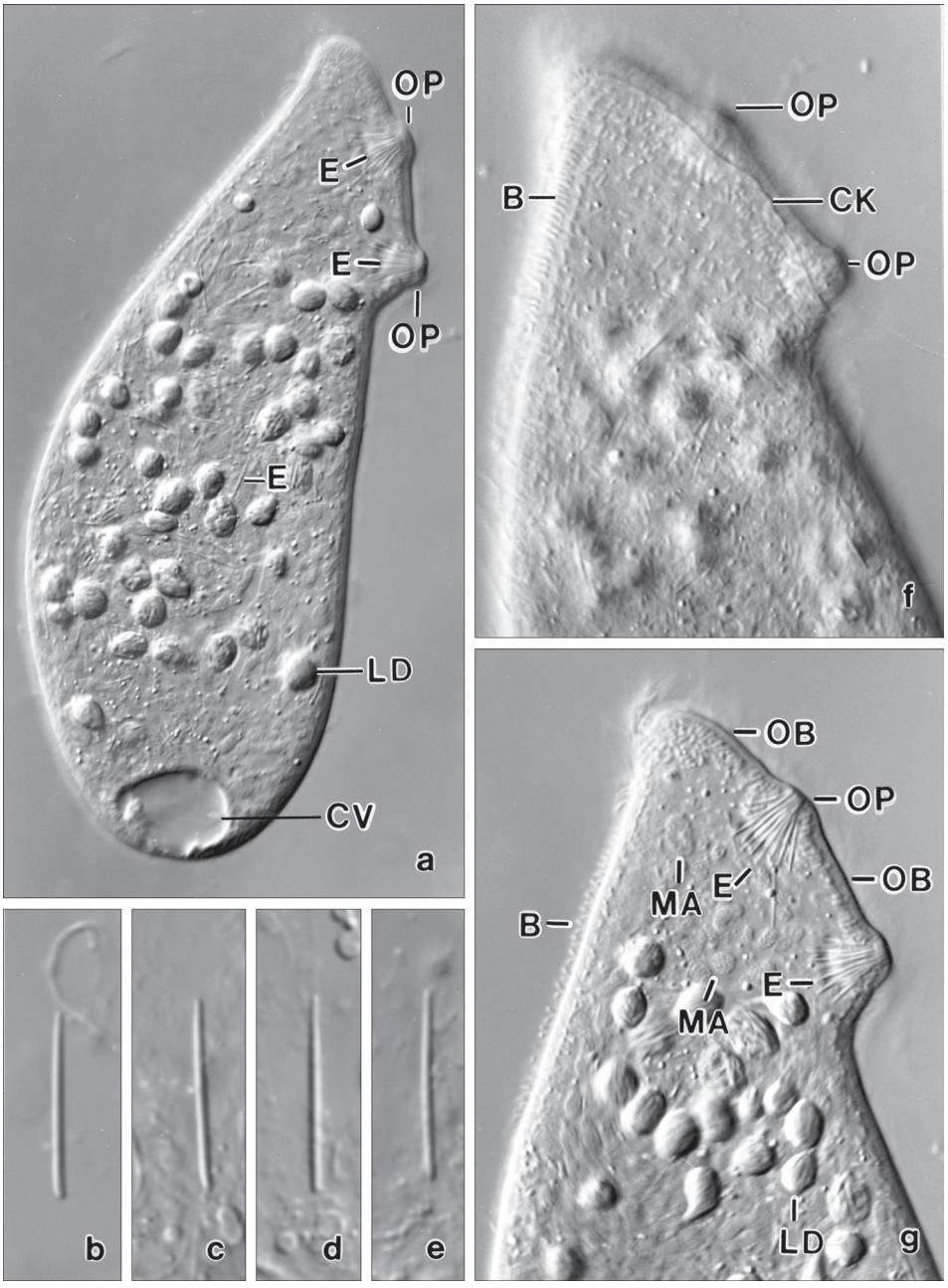

Fig. 6.11a–g *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of population from Vienna, Austria. From life, differential interference contrast). The specimens have two oral papillae and many macronucleus nodules. **a:** Right side view of a slightly squeezed specimen. **b–e:** The extrusomes are indistinctly acicular and about 12 × 0.7 μm in size. **f, g:** Anterior body portion in right side view showing the dorsal brush and the extrusomes in the oral papillae. B – dorsal brush, CK – circumoral kinety, CV – contractile vacuole, E – extrusomes, LD – lipid droplets, MA – macronucleus, OB – oral bulge, OP – oral papillae.

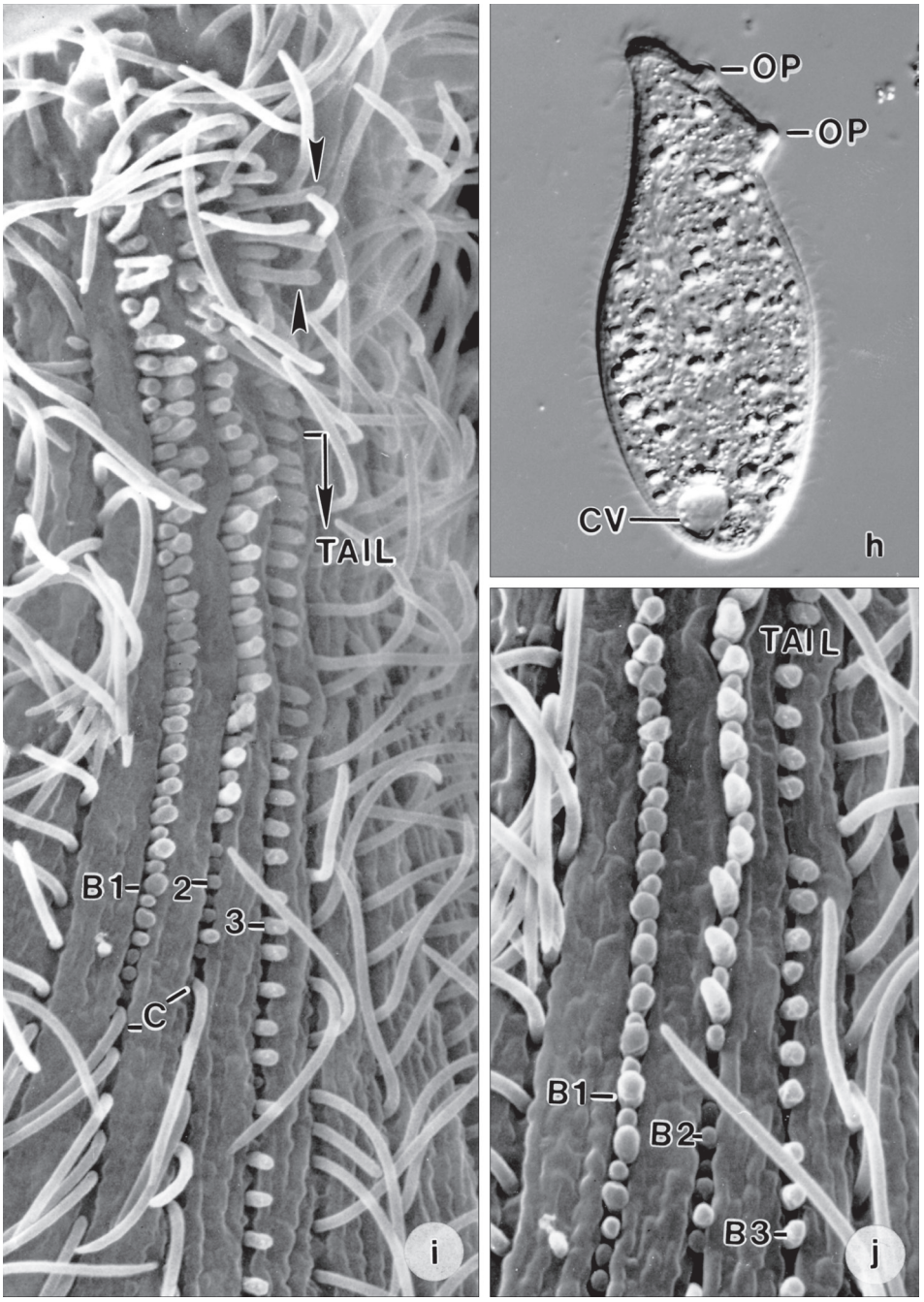

Fig. 6.11h–j *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of specimens from Salzburg [h] and Vienna [i, j]. h, from life; i, j, SEM). **h:** Right side view of a slightly squeezed specimen. **i, j:** Brush rows 2 and 3 have a cluster of elongated bristles subapically (arrowheads in i). Note the long, monokinetidal tail (arrow in i) of brush row 3. B1–3 – dorsal brush rows, C – ordinary somatic cilia, CV – contractile vacuole, OP – oral papillae.

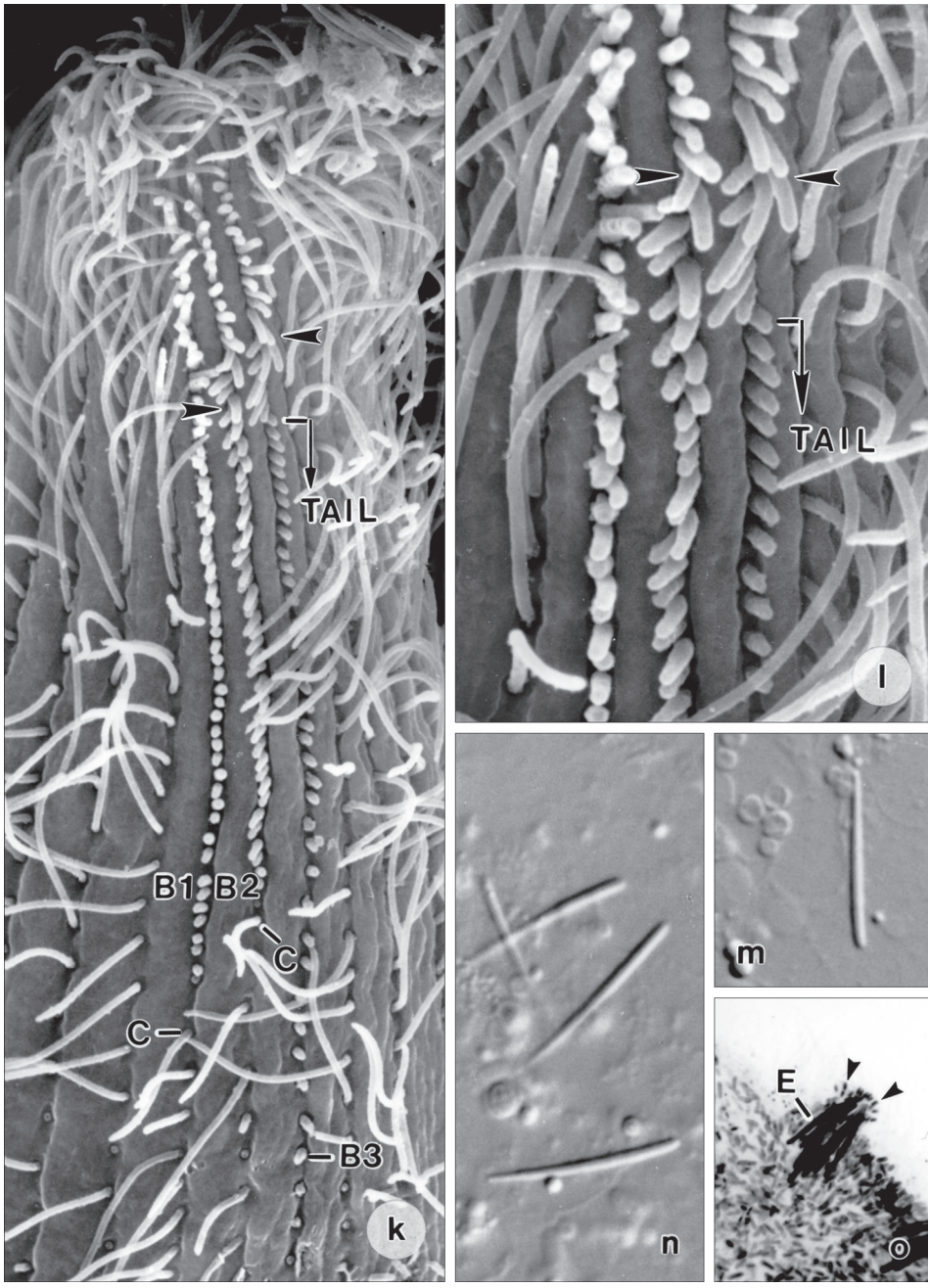

Fig. 6.11k–o *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals of specimens from Vienna [k, l] and South Africa [m–o]. k, l, SEM; m, n, from life; o, silver carbonate preparation). **k, l:** Overview of dorsal brush showing a cluster of elongated bristles (arrowheads) in subapical region of rows 2 and 3. Note the long, monokinetidal bristle tail of brush row 3 (arrows). **m–o:** Long and short (arrowheads in o) extrusomes. B1–3 – dorsal brush rows, C – ordinary somatic cilia, E – extrusomes.

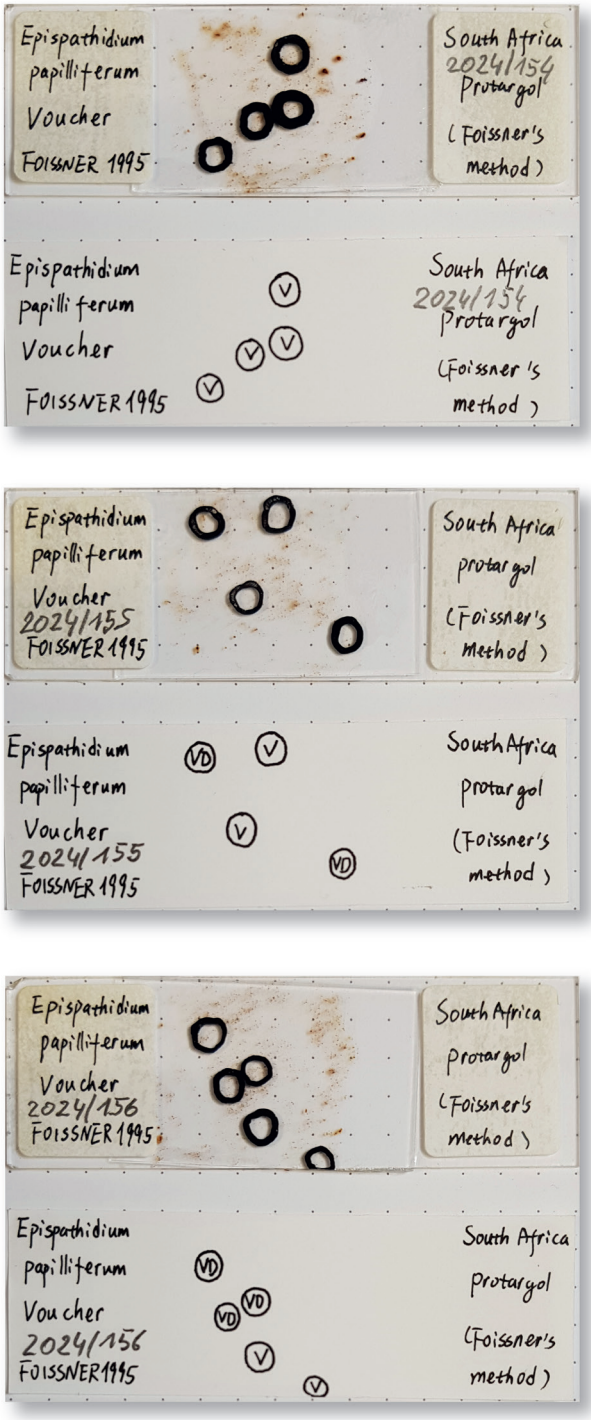

Fig. 6.12a–f *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. Protargol slides). **a–f:** Slides (a, c, e) and protocols (b, d, f) of voucher specimens (V) and voucher specimens drawn (VD) of population from South Africa. Accession numbers (LI): 2024/154, 155, 156.

papillae. Macronucleus usually composed of several to many scattered nodules, rarely of a tortuous strand. One or two types of extrusomes: type I studded in oral papillae and scattered in cytoplasm, rod-shaped to acicular and 10–13 × 0.5–1.0 µm in size; type II, if present, rod-shaped and fine, 1.5–2.0 µm long. About 28 ciliary rows, three of them anteriorly modified to rather distinct dorsal brush rows occupying about one third of body length; brush dikinetids obliquely arranged in anterior three quarters, at least in row 1; row 3 distinctly shortened.

Remarks (Table 6.5): *Epispathidium papilliferum* is unique in having oral bulge papillae, a striking feature degrading other main characteristics, such as the macronucleus pattern and extrusome shape. Kahl (1930a) found a single, "abnormal" specimen of *Epispathidium papilliferum* with only two papillae (dorsal one lacking) in Germany (Fig. 6.7g) and many "typical"

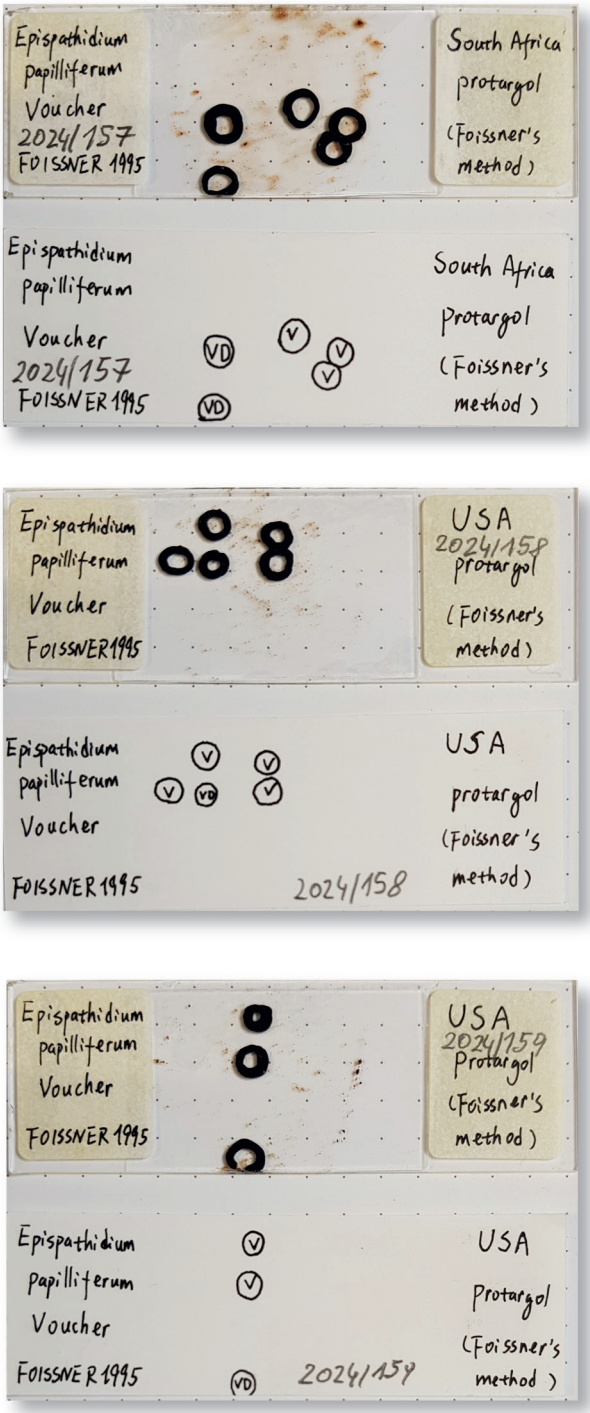

Fig. 6.12g–l *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. Protargol slides). **g, h:** Slide (g) and protocol (h) of voucher specimens (V) and voucher specimens drawn (VD) of population from South Africa. Accession number (LI): 2024/157. **i–l:** Slides (i, k) and protocols (j, l) of voucher specimens (V) and voucher specimens drawn (VD) of population from USA. Accession numbers (LI): 2024/158, 159.

specimens with three papillae in Zillertal in Austria (Fig. 6.7i). Foissner (1984) observed two populations, viz., one invariably with two papillae in Germany (Fig. 6.7a–e, j, k) and another with three warts in Austria (Fig. 6.7f). Further populations show that this species has either two papillae (e.g., in a Salzburg and a Vienna population) or three (e.g., in another Vienna population and in specimens from Berlin, Finland, and the USA; Fig. 6.11h–l). Further, both types may occur together, as shown by the South African and Slovakian populations (Table 6.5; Tirjaková et al. 2002, p. 238, their Fig. 5). Jang et al. (2017, their Fig. 7a–i) also found specimens with two or three papillae in the same population.

All these and several other populations observed over the years have scattered macronucleus nodules accompanied by either two oral papillae or three. Thus,

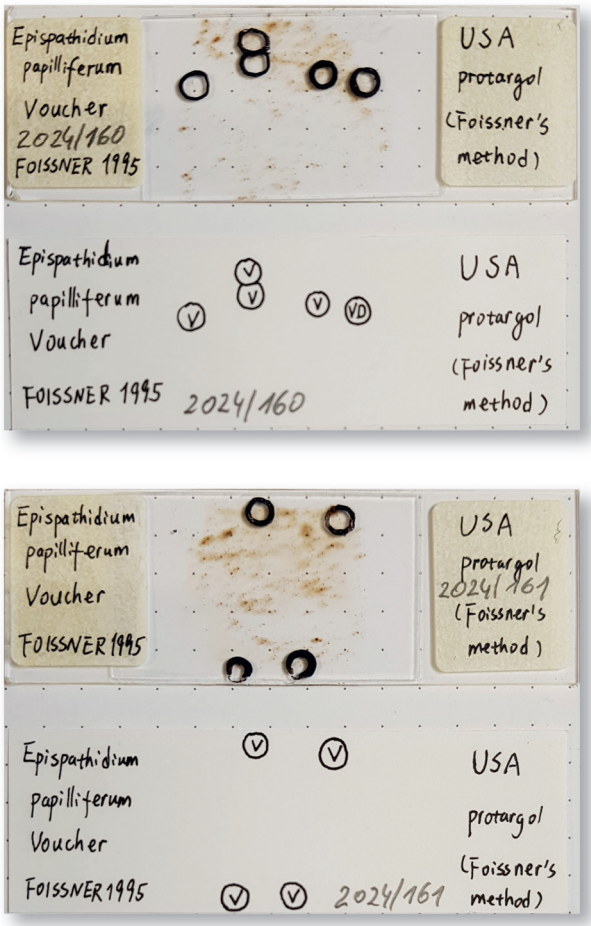

Fig. 6.12m–p *Epispathidium papilliferum* (Kahl, 1930) Foissner, 1984 (originals. Protargol slides). **m–p:** Slides (m, o) and protocols (n, p) of voucher specimens (V) and voucher specimens drawn (VD) of population from USA. Accession numbers (LI): 2024/160, 161.

the South African population, where 127 out of 172 specimens have a long, more or less moniliform macronucleus strand, is a real exception. This ratio and the lack of transition stages show that the pattern is not caused by post-divisional macronucleus processes. In addition, there is variation in the shape (rod-shaped to acicular) and types (one or two) of extrusomes (Table 6.5). However, all recent observations suggest a rather similar, acicular or slightly acicular shape of the long (main) extrusomes. Likewise, we cannot exclude the occurrence of a second extrusome type, viz. short, indistinct rods, in most populations. However, the new observations on Vienna specimens show that there are indeed populations without the second type, indicating that this feature is variable and should not be over-emphasized at the present state of knowledge. All these observations indicate distinct subpopulations, probably (sub)species in status nascendi.

Epispathidium papilliferum is either a species with considerable variability, a species just splitting into several subspecies, or a complex of three different species/subspecies, whose diagnostic features, however, do not match. Thus, it would be premature to split it at the present state of knowledge. Clearly, *Epispathidium papilliferum* is a challenge for gene sequence studies which show that the populations studied so far are not closely related in molecular trees (Jang et al. 2017, their Fig. 16; Pomahač et al. 2023, their Fig. 11; see also note on p. 211).

Vuxanovici (1959, p. 323, Plansa VI, Fig. 42) described the species *Spathidium papillatum*. However, the extrusome warts of this species are not on the oral bulge, but on the anterior body half. Perhaps it is related to *Legendrea* Fauré-Fremiet, 1908 which also has the warts (tentacles) on the body (Pomahač et al. 2023, Weiss et al. 2022).

Jankowski (2007) fixed *Spathidium papilliferum* as type species of *Vartospathidium* because he considered the extrusome warts on the oral bulge as especially important. More detailed data are needed to support or disprove the hypotheses if the current species belongs to *Spathidium*, *Epispathidium*, or to *Vartospathidium*. In the tree published by Pomahač et al. (2023), *Spathidium papilliferum* sequences are distributed throughout the spathidiid tree.

Description: The description is based on the literature cited above and four further populations studied over the years. The individuals from South Africa ((Fig. 6.8a–v, 6.10a–j, l, m, o, p, 6.11m–o) and North America (USA; Fig. 6.9a–d, f–l, 6.10k, n, q) were investigated in detail, while the specimens from Vienna (Fif. 6.9e, 6.11a–g, k, l) and Salzburg (Fig. 6.11h) were identified routinely in vivo by the main features, namely, the oral papillae, extrusomes, and macronucleus pattern. Interestingly, three types were observed with respect to the oral papillae and macronucleus pattern (Table 6.5): (i) three papillae and many scattered macronucleus nodules in the USA population; (ii) two papillae and many scattered macronucleus nodules in the Vienna and Salzburg populations; and (iii) a combination of (i) and (ii) in the South African specimens, where 172 cells were analysed: 45 had two papillae and many scattered macronucleus nodules (SAN-type, South African Nodules), as usual, while 127 had three papillae and a single, more or less moniliform macronucleus strand (SAS-type, South African Strand). The proportion of the SAS-type to SAN-type specimens was about 2:1 ten days after wetting the soil sample and increased to 3:1 on day 20. The two types of South African specimens match well each other and other populations in almost all other main features. Thus, conspecificity is likely and the population with the single macronucleus included in the following description. For detailed description of two Korean populations, see Jang et al. (2017); their data are not included in the text below, but they agree well with the compilation below.

Body size and length:width ratio of individual populations moderately variable in vivo and protargol preparations (Table 6.4): in vivo 170–260 μm with a length:width ratio of 4:1 in type population (Kahl 1930a, b); 100–200 × 30–60 μm in vivo, 115–170 × 27–42 μm, on average 138 × 34 μm in protargol preparations of the German population described by Foissner (1984); 109–144 × 31–66 μm, on average 126 × 45 μm in protargol-prepared specimens from the USA; 114–161 × 22–42 μm, usually about 139 × 31 μm in protargol-prepared South African population. Taking into account some shrinkage and previous data, a usual in vivo size of 140–170 × 30–50 μm and a length: width ratio of about 4:1 can be estimated. Body narrowly spatulate with oblique anterior (oral) end highly conspicuous due to the two or three extrusome papillae; neck ordinarily distinct, trunk widest behind mid-body, posterior end broadly rounded; laterally flattened up to 3:1, especially in hyaline anterior body portion (Fig. 6.7a–c, g–i, 6.8a, d–g, 6.9a, f, g, 6.10a, k–m, 6.11a, h). Macronucleus divided into seven to about 100 scattered nodules, rarely in a tortuous, more or less moniliform strand, viz., in the South African specimens (see above). Individual nodules ellipsoid to globular, contain several globular nucleoli. Micronuclei scattered in middle body portion, globular and about 2 μm across, clearly recognizable only in SAS-type of South African specimens (Fig. 6.7d, f, g, i, 6.8m–r, t, 6.9j–l, 6.10a, k–m, o; Tables 6.4, 6.5). Contractile vacuole in rear body end, several excretory pores in pole area. One or two types of extrusomes in the oral papillae and scattered in cytoplasm, type I lacking in oral bulge between papillae (Fig. 6.7a, b, g, i, 6.8a, i, j, r, 6.9a, d, e, k, 6.10a–d, f–j, 6.11b–e, m–o; Table

6.5). Type I extrusomes conspicuous, rod-shaped and about 13 µm long according to Kahl (1930a); rod-shaped and 10.0–12.0 × 0.5 µm in German population described by Foissner (1984); indistinctly acicular and about 12.0 × 0.7 µm in a population from Vienna (Fig. 6.11a–g); almost rod-shaped to acicular and about 10.0 × 0.7 µm in Salzburg population; almost acicular, slightly curved, and about 10 × 1 µm in USA population; almost rod-shaped and about 11 µm long in SAN-type, while acicular, straight to slightly curved, and 12–13 × 1 µm in SAS-type of South African specimens; do not impregnate with protargol. Type II, if present, easily overlooked because minute, but definitely lacking in a Vienna population; fine and rod-shaped, about 2 µm long in USA population, and 1.5 × 0.2 µm in SAS-type of South African specimens. Exploded type I extrusomes of typical toxicyst structure, about 25–45 µm long in vivo. Developing extrusomes scattered in cytoplasm, fusiform and 8–11 µm long, frequently impregnate with protargol in SAS-type of South African specimens. Cortex flexible and about 2 µm thick, contains loosely spaced granules about 0.2 µm across in South African specimens; frequently impregnate with protargol in SAS-type, but never in SAN-type contained in same preparations (Fig. 6.8c, 6.9c). Cytoplasm colourless, often packed with lipid droplets up to 10 µm across, extrusomes, and 7–15 µm-sized food vacuoles containing definitely bacteria in some Vienna specimens; remnants of ciliate prey (*Frontonia depressa*) in USA population, where a specimen with widely opened mouth was found in the preparations, showing that even the papillae split when the bulge opens (Fig. 6.9k, l, 6.10n); and mainly flagellates in SAS-type of South African specimens, which frequently have a subterminal vacuole with indigestible debris. Glides slowly on microscope slide (German population) or swims rather rapidly (South African specimens).

Somatic cilia about 12 µm long in vivo and ordinarily spaced (2.0–2.5 µm on average), except for anterior end of rows, where closely spaced cilia produce distinct kinetofragments, forming, together with the circumoral cilia, a conspicuous corona; arranged in 22–36, usually near 28 equidistant, bipolar rows anteriorly arranged in a pronounced *Epispathidium* pattern, which is, unexpectedly, maintained even in a specimen with widely opened mouth (Fig. 6.9k, l, 6.10n; Table 6.4). Dorsal brush dikinetidal and three-rowed, rather distinct because occupying an average of 27–39% of body length and bristles up to 4–6 µm long in vivo; all rows commence with some ordinary cilia anteriorly, rows 1 and 2 end at nearly same level and continue with ordinary cilia posteriorly. Row 1 composed of an average of 35–56 densely spaced dikinetids obliquely arranged in the anterior three quarters, occasionally mixed with some monokinetids posteriorly in SAS-type of South African specimens; row 2 composed of an average of 28–37 dikinetids comparatively loosely spaced in posterior half and anteriorly slanted in German specimens; row 3 distinctly shorter than rows 1 and 2, composed of an average of only 9–20 dikinetids anteriorly slanted in German specimens, followed by a monokinetidal tail of rod-shaped, about 2 µm long bristles extending to midbody. Brush bristles slightly clavate, decreasing in length from 5–6 µm anteriorly to 2.5–3.0 µm posteriorly in rows 1 and 2 of South African specimens both in vivo and protargol preparations; up to 5 µm long anteriorly, decreasing to about 3 µm posteriorly in a Salzburg (Gaisberg area) population, where the anterior bristles of the dikinetids are only half as long as the posterior ones. Details of the dorsal brush could be studied in SEM-micrographs from two specimens of a population from the surroundings of Vienna (Fig. 6.11i–l): (i) the brush rows extend in distinct furrows separated by broad, flat ridges; (ii) the narrowly spaced and

continued on p. 194

Table 6.4 Morphometric data on several populations of *Epispathidium papilliferum*. SAS, SAN: South African population (original data) 10 d after wetting the sample, separated in specimens with macronucleus either in a tortuous strand (SAS) or in many scattered nodules (SAN); USA: North American population (original data); GER: population from Rauisch-Holzhausen, Germany (from Foissner 1984)[a]

Characteristic	Pop	Mean	M	SD	SE	CV	Min	Max	n
Body, length	SAS	142.2	142.0	9.2	2.1	6.5	125.0	161.0	19
	SAN	136.0	136.0	12.0	2.7	8.8	114.0	158.0	19
	USA	126.1	124.0	10.1	2.2	8.0	109.0	144.0	21
	GER	137.9	140.0	19.4	7.4	14.1	115.0	170.0	7
Body, width	SAS	32.1	30.0	4.6	1.1	14.5	26.0	42.0	19
	SAN	30.3	30.0	5.2	1.2	17.1	22.0	40.0	19
	USA	45.0	46.0	8.7	1.9	19.2	31.0	66.0	21
	GER	33.6	35.0	5.9	2.6	17.6	27.0	42.0	5
Body length:width, ratio	SAS	4.5	4.5	0.5	0.1	11.8	3.5	5.3	19
	SAN	4.6	4.6	0.6	0.1	12.7	3.7	5.9	19
	USA	2.9	2.8	0.5	0.1	18.1	2.2	4.0	21
	GER			about 4.1					
Oral bulge, length	SAS	37.5	36.0	4.9	1.1	13.2	30.0	52.0	19
	SAN	38.0	38.4	4.5	1.0	11.9	30.0	46.0	19
	USA	27.6	26.4	3.8	0.8	13.8	22.0	36.0	21
	GER	36.6	38.0	3.4	1.5	9.4	31.0	40.0	5
Oral bulge, width between papillae	SAS	2.3	2.3	–	–	–	2.0	2.6	4
	SAN	2.6	2.5	–	–	–	2.4	3.0	5
	USA	2.0	2.0	–	–	–	1.8	2.4	5
Oral bulge, height between papillae	SAS	2.3	2.2	–	–	–	2.0	2.6	19
	SAN	2.3	2.4	–	–	–	2.0	2.4	19
	USA	2.6	2.4	–	–	–	1.8	4.2	19
Oral papillae, number	SAS	3.0	3.0	0.0	0.0	0.0	3.0	3.0	19
	SAN	2.0	2.0	0.0	0.0	0.0	2.0	2.0	19
	USA	3.0	3.0	0.0	0.0	0.0	3.0	3.0	21
	GER	2.0	2.0	0.0	0.0	0.0	2.0	2.0	15
Oral papillae, across	SAS	4.0	4.0	–	–	–	3.8	4.2	4
	SAN	4.6	4.7	–	–	–	4.2	4.8	5
	USA	3.5	3.5	–	–	–	3.4	3.6	5
Oral papillae, height	SAS	3.9	3.6	0.7	0.2	18.7	3.0	6.0	19
	SAN	4.7	4.8	0.9	0.2	18.9	3.6	6.0	19
	USA	3.7	3.6	0.5	0.1	13.9	2.4	4.8	21
Circumoral kinety to last dikinetid of brush row 1, distance	SAS	44.6	46.0	4.9	1.5	10.9	37.0	52.0	11
	SAN	52.6	53.0	4.7	1.8	8.9	47.0	60.0	7
	USA	33.9	34.0	3.3	0.8	9.7	30.0	41.0	19
Circumoral kinety to last dikinetid of brush row 2, distance	SAS	44.0	42.0	4.6	1.4	10.5	37.0	52.0	11
	SAN	50.1	50.0	3.7	1.4	7.4	46.0	56.0	7
	USA	32.3	34.0	3.6	0.8	11.0	25.0	41.0	19
	GER	39.2	40.0	3.0	1.3	7.5	35.0	43.0	5
Circumoral kinety to last dikinetid of brush row 3, distance	SAS	11.9	12.0	3.3	1.0	28.0	7.0	18.0	11
	SAN	13.7	13.0	3.0	1.1	21.9	11.0	19.0	7
	USA	13.3	12.0	5.3	1.2	39.9	7.0	26.0	19
	GER	13.8	10.0	8.3	3.7	59.9	8.0	28.0	5
Macronucleus figure, length	SAS	43.9	43.0	9.2	2.1	20.9	26.0	62.0	19

Table 6.4 Continued

Characteristic	Pop	Mean	M	SD	SE	CV	Min	Max	n
Macronucleus figure, length	SAN	78.8	82.0	8.7	2.0	11.0	56.0	92.0	19
	USA	79.5	77.0	9.1	2.0	11.4	66.0	100.0	21
Anterior body end to anteriormost macronucleus nodule, distance	SAS	69.4	68.0	15.1	3.5	21.7	47.0	94.0	19
	SAN	39.3	41.0	4.6	1.1	11.8	28.0	46.0	19
	USA	30.4	30.0	3.7	0.8	12.1	24.0	38.0	21
Macronucleus, length (spread)	SAS	90.8	90.0	–	–	–	–	118.0	19
Macronucleus nodules, length	SAN	7.5	7.2	1.7	0.4	22.5	5.0	11.0	19
	USA	8.0	7.2	2.3	0.5	28.2	5.0	13.0	21
	GER	5.3	5.6	0.9	0.3	16.1	4.0	7.0	11
Macronucleus, width	SAS	5.5	5.0	0.9	0.2	15.6	4.8	8.0	19
Macronucleus nodules, width	SAN	2.9	2.4	0.8	0.2	27.9	2.2	4.8	19
	USA	2.8	2.6	0.7	0.2	24.7	1.8	4.5	21
	GER	2.2	2.0	0.5	0.1	21.4	1.6	2.8	11
Macronuclei, number	SAS	1.0	1.0	0.0	0.0	0.0	1.0	1.0	19
	SAN	77.4	79.0	14.0	4.9	18.1	54.0	98.0	8
	USA	59.6	58.0	12.1	2.8	20.4	35.0	80.0	19
	GER	about 100 nodules							
Micronuclei, diameter	SAS	1.8	1.8	0.3	0.1	15.3	1.5	2.4	19
Micronuclei, number	SAS	13.6	14.0	3.2	0.7	23.8	9.0	21.0	19
Basal bodies in a right-side kinety, number	SAS	58.2	56.0	6.4	2.9	11.0	52.0	68.0	5
	SAN	58.8	58.5	7.3	2.1	12.5	50.0	75.0	12
	USA	65.8	65.0	11.2	2.9	17.0	50.0	83.0	15
	GER	78.0	80.0	16.1	7.2	20.6	55.0	100.0	5
Somatic kineties, number	SAS	29.8	28.0	3.7	1.1	12.4	25.0	35.0	12
	SAN	28.1	27.0	2.3	0.6	8.2	25.0	33.0	17
	USA	24.4	25.0	1.4	0.3	5.6	22.0	28.0	21
	GER	33.3	33.0	1.5	0.6	4.5	32.0	36.0	6
Dorsal brush rows, number	SAS	3.0	3.0	0.0	0.0	0.0	3.0	3.0	19
	SAN	3.0	3.0	0.0	0.0	0.0	3.0	3.0	19
	USA	3.0	3.0	0.0	0.0	0.0	3.0	3.0	21
	GER	3.0	3.0	0.0	0.0	0.0	3.0	3.0	5
Dikinetids in brush row 1, number	SAS	37.0	38.0	3.7	1.1	10.0	30.0	41.0	11
	SAN	43.6	44.0	3.2	1.2	7.2	39.0	47.0	7
	USA	34.5	35.0	4.5	1.0	13.0	24.0	41.0	20
	GER[b]	56.0	–	–	–	–	54.0	58.0	2
Dikinetids in brush row 2, number	SAS	29.0	29.0	2.9	0.9	10.1	25.0	34.0	11
	SAN	31.0	31.0	4.2	1.6	13.7	23.0	36.0	7
	USA	27.7	28.0	2.8	0.6	10.2	22.0	32.0	20
	GER[b]	37.0	–	–	–	–	35.0	39.0	2
Dikinetids in brush row 3, number	SAS	9.7	9.0	3.1	0.9	32.2	5.0	16.0	11
	SAN	11.6	11.0	2.1	0.8	18.6	9.0	14.0	7
	USA	10.1	9.0	3.1	0.7	30.5	7.0	17.0	20
	GER[b]	20.0	–	–	–	–	19.0	21.0	2

[a] Data based on mounted, protargol-prepared (Foissner's method), and randomly selected specimens from non-flooded Petri dish cultures. Measurements in μm. For abbreviations, see footnote a of Table 6.3.

[b] From figures.

Table 6.5 Comparison of some main features in *Epispathidium papilliferum* populations[a]

Population[b]	P	Macronucleus	Type I extrusomes, shape and length	n
Zillertal (from Kahl 1930a)	3	7–10 nodules	rod-shaped, 13 µm	many?
Mittenwald (from Kahl 1930a)	2	7 nodules	?	?
Gießen (from Foissner 1984)	2	about 100 nodules	rod-shaped, 10–12 µm	15
Seekirchen (from Foissner 1984)	3	many nodules	?	few
Vienna[c]	2	many nodules	indistinctly acicular, 12 µm	several
Salzburg[d]	2	50–100 nodules	rod-shaped, acicular, 10 µm	several
Pennsylvania	3	35–80 nodules	almost acicular, 10 µm	50
South Africa (SAN-type)	2	54–98 nodules	almost rod-shaped, 11 µm	45
South Africa (SAS-type)	3	a tortuous strand	acicular, 12–13 µm	127

[a] Abbreviations: n – number of specimens analysed, P – number of papillae.
[b] For detailed description of sample sites, see occurrence and ecology. Data in lines 5 ("Vienna") to 9 ("South Africa (SAS-type") are original observations.
[c] Floodplain near Vienna (Austria) in 2002.
[d] Beech forest in the Gaisberg area (Salzburg, Austria) in 1997.

obliquely arranged dikinetids in the anterior portion of rows 1 and 2 are also prominent in the SEM, however, only in one of the two specimens: the brush furrows are distinctly widened in this region and the brush rows appear doubled due to the zigzagging kinetids; (iii) a cluster of distinctly elongated, slightly clavate bristles occurs in rows 2 and 3 at level of transition zone of dikinetidal and monokinetidal portion of row 3; (iv) the bristles are slightly clavate in one specimen and rod-shaped in the other; (v) the anterior bristle of the dikinetids is slightly shorter than the posterior one in one of the two specimens; (vi) the bristles of the monokinetidal tail of row 3 are rod-shaped.

Oral bulge slanted by 30–60°, about as long as widest trunk region, 4–6 µm high in vivo, very conspicuous because bearing two (invariably ventral and middle) or three (ventral, middle, and dorsal) extrusome papillae; in frontal view oblong with base of papillae slightly inflated; surface bent dorsally from middle to dorsal end in type population and USA specimens, slightly curved or sigmoidal in German, Austrian (Foissner 1984), and South African specimens; individual papillae with minute concavity apically, 3.5–4.6 µm across and 3.7–4.7 µm high in protargol preparations. Circumoral kinety of same shape as oral bulge when viewed frontally, continuous, and distinctly separate from ciliary rows; composed of dikinetids each associated with an oral basket rod, a fibre extending into bulge and to papillae centre, and a cilium about 12 µm long in South African, 15 µm in USA, and 18 µm in German specimens. Oral basket rather distinct, composed of nematodesma bundles originating from circumoral kinety (Fig. 6.7j, 6.8b, k, l, q–t, 6.9h, i, 6.10a, k–m, p, q, 6.11a, f–h; Tables 6.4, 6.5).

Resting cyst: Jang et al. (2017, p. 954) described the resting cyst (about 40–45 µm in diameter, wall smooth) of a Korean population.

Molecular biology: 18S rRNA gene analyses were performed by Strüder-Kypke et al. (2006; GenBank accession numbers DQ411857, isolate A; DQ411858, isolate B; both isolates are from a population from a forest soil from the Müllerboden, Austria; details on

sample site, see Foissner et al. 2005, p. 619; identification by W. Foissner). For Korean sequences KY556645 and KY556649, see Jang et al. (2017, p. 941). As expected, the two Austrian sequences cluster together because they are from the same population. However, the Korean sequences are widely separated from each other and the Austrian population (e.g., Chi et al. 2022, p. 3/15; Jang et al. 2017, p. 969; Jang et al. 2022, p. 7/11). Whether these different positions in molecular trees reflect the true relationships (different species with oral bulge papillae due to convergent evolution) or are due to methodological problems must be analyzed by detailed studies based on reliable identified populations.

Occurrence and ecology: The data below show that *Epispathidium papilliferum* occurs in all main biogeographic regions, except of Antarctica and Palaeotropis (Foissner 1998, p. 203; for a record from South Africa, see below). It is a rather common, but rarely abundant terrestrial species which probably prefers acidic to circumneutral environments. No records are known from saline habitats.

Kahl (1930a) found this species in two sites, which are, however, only about 50 km away from each other. At first, he found a single specimen (Fig. 6.7g, h) in lime-rock moss from Mittenwald, a municipality in the district of Garmisch-Partenkirchen, Upper Bavaria (this is the southern portion of Bavaria), Germany; later he found two further specimens likewise with only two warts on the same locality (Kahl 1930a, p. 387). The second population (Fig. 6.7i) showed high abundance and was isolated from moss from the Zillertal (the northern end of this valley is about at 47.40°N 11.83°E; Tyrol, Austria); in the legend to Fig. 6.7i, Kahl (1930a, p. 390) wrote "*Sp. papilliferum* typische Form" indicating that he considered the Austrian specimens as type population.

Foissner (1984, p. 6, 7) found a population each in wall moss from the village Rauisch-Holzhausen (about 50.758°N 08.882°E) near Gießen, Germany (rare; Table 6.4) and in leaf litter from a mixed forest near the townlet Seekirchen (about 47.89°N 13.12°E), Salzburg, Austria (very rare; Fig. 6.7g; Foissner 1984, p. 84). Foissner W. also observed it in soil from a beech forest of the Gaisberg area (local mountain east of city of Salzburg, Austria), at pH 5.0–6.7 (Fig. 6.11h); soil/litter from a mixed forest from the surroundings of Berlin, Germany; moss from a coniferous forest in Lapland, Finland, at pH 4.3; and soil from deciduous and coniferous forests near Vienna, Austria, at pH 4.5–7.4 (e.g., Foissner et al. 2005, p. 627; unpublished observations). In North America, *Epispathidium papilliferum* occurred in leaf litter (pH 5) of American Helmlock trees at the trailhead of the Gifford Pinchot trail system in the Lackawanna State Forest, Lackawanna County (about 41.45°N 75.61°W), Pennsylvania, USA (sample likely provided by A. Berthold, original observations). In South Africa, it occurred in mossy bark (pH 6.3) mainly from "Hard Pear" (*Olinia ventosa*) trees in the Kirstenbosch Botanical Garden, viz., at the trailhead (about 33.987°S 18.431°E) of the Skeleton Gorge trail to the Table Mountain (original observations).

Tirjaková et al. (2002) found *Epispathidium papilliferum* in one (leaf-litter) out of 15 samples from acidic (pH ~4) oak-hornbeam forests of western Slovakia. The Korean populations studied by Jang et al. (2017) are from leaf litter and soil from Jungjok mountain, Joil-ri, Samdong-myeon, Ulju-gun, Ulsan (35.456°N 129.136°E) and from leaf litter and soil from the Ssanggyesa temple, Sacheon-ri, Uisin-myeon, Jeollanam-do (34.468°N 126.308°E).

There exist several records not substantiated by morphological data (however, due to the papillae a confusion with other species is very unlikely) showing that *Epispathidium*

papilliferum prefers terrestrial habitats[15]: dry moss from Bavaria (surroundings of the city of Erlangen, Gößweinstein, or Seeon), Germany (Wenzel 1953, p. 71, 79); *Melico-Fagetum* soil from Erminger forest (about 48.38°N 09.88°E), Germany (Lehle 1989, p. 133, 140); dry moss in Slovakia (Tirjaková & Matis 1987, p. 10; Matis et al. 1996, p. 11); leaf litter and moss (pH 5.1) in the Brisbane Water National Park, Australia (Blatterer & Foissner 1988, p. 7); litter and roots (pH 5.7) from an *Eucalyptus* forest in the Belair National Park near Adelaide, Australia (Blatterer & Foissner 1988, p. 7); soil (pH 7.2) from Hawaii, together with *Arcuospathidium cultriforme cultriforme* (Penard, 1922) Foissner, 1984 (original observation)[16]; South America (Foissner 1998, p. 203; without details on site).

Only Grabacka (1977, p. 380) recorded *Epispathidium papilliferum* from freshwater, viz., the upper 5 mm mud layer of a Polish fishpond, which received sugar factory wastes. Details were not provided; thus, misidentification cannot be excluded, though this is not very likely considering the highly characteristic extrusome papillae. Biomass of 10^6 specimens about 135 mg (Foissner 1998, p. 203).

Brief review of other species assigned to *Epispathidium* Foissner, 1984[17]

Epispathidium terricola Foissner, 1987

1984 ***Spathidium muscicola* Kahl, 1930** – Berger, Foissner & Adam, Zool. Jb. Syst. 111: 358, Abb. 48–51, Tabelle 6 (description of Austrian population; likely a misidentification, see nomenclature and remarks).

1987 ***Epispathidium terricola* nov. spec.**[18] – Foissner, Sber. öst. Akad. Wiss., Mathematisch-naturwissenschaftliche Klasse, Abt. I 195 (year 1986): 234, Abb. 11a–i, 12, Tabelle 2 (original description; the slide containing the holotype [accession number 1988/151; holotype not designated in original description] and one paratype slide [1988/152] have been deposited in the Biology Centre of the Upper Austrian Museum in Linz [LI]; see Aescht 2008, p. 182).

2006 ***Epispathidium terricola* Foissner, 1986** – Alekperov & Sadikhova, Turk. J. Zool. 30: 400, Fig. 2A, B (description of population from Azerbaijan using wet silver nitrate preparation method; site were voucher slides deposited not mentioned).

2007 ***Spathidium terricola*** – Foissner & Xu, Monogr. boil. 81: 38 (non-formal combination with *Spathidium*, see remarks).

2016 ***Epispathidium terricola* Foissner, 1987** – Rajter & Vdačný, Zool. Scr. 45: 202 (gene sequence analyses of 2 terrestrial populations from Slovakia based on non-clonal specimens identified in vivo).

2017 ***Spathidium terricola* (Foissner, 1987) comb. nov.** – Jang, Vdačný, Shazib & Shin, J. nat. Hist. 51: 971 (combination with *Spathidium*).

Nomenclature: No etymology has been provided in the original description. For derivation of the species-group name *terricola*, see Berger (2011, p. 419). Voucher slides of the population studied by Berger et al. (1984) have not been found in the Biology Centre of the Upper

[15] Note by H. Berger: W. Foissner wrote this part in the early 2000s. Thus, younger records are not included. I refrained from a detailed update of the records not substantiated by morphological data for time reasons.

[16] Note by H. Berger: Likely this sample is from the "Hawaii, Big Island site 39" sample mentioned by Foissner & Xu (2007, p. 236).

[17] Written by H. Berger. For figures, see individual papers mentioned in list of synonyms.

[18] Foissner (1987) provided the following diagnosis: "In vivo etwa 120–160 × 35–60 µm großes, spathidiformes, farbloses *Epispathidium* mit ungefähr 40 µm langen Extrusomen, bandförmigem Makronucleus und durchschnittlich 39 Somakineten. Verbreitet in edaphischen Biotopen."

Austrian Museum in Linz (LI). Further, they could not be localized in the private collections of W. Foissner and H. Berger in Salzburg within a reasonable time.

Diagnosis (from Foissner 1987, slightly modified): Body size about 120–160 × 35–60 µm in vivo; body shape spathidiforme; body colourless; extrusomes about 40 µm long; macronucleus ribbon-like, serpentine; on average 30 somatic kineties. Common in terrestrial habitats.

Remarks: According to Foissner (1987, p. 237), *Spathidium muscicola* Kahl, 1930a (p. 377) sensu Berger et al. (1984) is likely a misidentification and belongs to the present species. Alekperov & Sadikhova (2006) incorrectly assumed that the type locality of *Epispathidium terricola* is in Austria; it is in Germany (see below). The specimens from Azerbaijan agree very well with those from the type locality, indicating that the identification is correct. Jang et al. (2017) transferred the present species to *Spathidium* because *Epispathidium* species do not group together in molecular trees. Since Foissner & Xu (2007, p. 38) made no formal combination, Jang et al. (2017) should be accepted as combining author when the species is classified in *Spathidium*.

Description: For description of type population, see Foissner (1987). For brief description of population from Austrian Alps, see Berger et al. (1984). For characterization of terrestrial population from Azerbaijan, see Alekperov & Sadikhova (2006).

Molecular biology: For gene sequence analyses of two populations, see Rajter & Vdačný (2016). Accession numbers of 18S rRNA in GenBank: KT246082, KT246083.

Occurrence and ecology: A very common species in all biogeographic regions in soil, less common in mosses (Foissner 1987, p. 237; 1998, p. 203; Foissner et al. 2005, p. 627). The type locality is soil (mainly the organic upper layer; pH ~3; sea level about 613 m) from a spruce forest (*Picea abies*) near the city of Ulm (48.39°N 09.98°E), Germany (Foissner 1987, p. 218, 234; for details, see Lehle 1989, p. 133, 140, sites U1NF, U1DF). Berger et al. (1984) found "*Spathidium muscicola*" in experimentally compacted alpine pasture soil (0–5 cm; 47.1458°N 13.0570°E) from the Schloßalm area, Bad Hofgastein, Salzburg, Austria (for details on this site, see Berger et al. 1985, p. 98, 107). Alekperov & Sadikhova (2006 p. 399) isolated the present species from soil from the Pirgulian State Reserve (about 40.77°N 48.50°E), near the city of Shemakha, eastern Azerbaijan. For details on sample sites of Slovakian populations, see Rajter & Vdačný (2016, p. 202). Biomass of 10^6 specimens about 101 mg (Foissner 1998, p. 203).

Epispathidium amphoriforme (Greeff, 1889) Foissner, 1984

(Fig. 6.3d, 6.13a–f)

1889 ***Spathidium amphoriforme*, nov. spec.** – Greeff, Sber. Ges. Beförd. ges. Naturw. Marburg 1888: 131 (original description, no illustration and no type material available).

1922 ***Spathidium amphoriforme* Greeff 1888** – Penard, Infusoires, p. 24, Fig. $17_{1, 2}$, $18_{3, 4}$ (redescription; no voucher material available).

1930 ***Spathidium amphoriforme* Greef (1888), Penard (1922)** – Kahl, Arch. Protistenk. 70: 377, Fig. 7e (description of Bavarian population; no voucher material available, incorrect spelling of Greeff).

1930 ***Spathidium amphoriforme* var. *rectitoratum* var. n.** – Kahl, Arch Protistenk. 70: 380, Fig. 9q (Fig. 6.3d; original description of variety; no type material available).

1930 ***Spathidium amphoriforme* Greef (1888) – Penard (1922)** – Kahl, Tierwelt Dtl. 18: 166, Fig. 24_6 (revision of ciliates; incorrect spelling of Greeff).

1930 ***Spathidium amphoriforme* var. *rectitoratum* Kahl, 1930** – Kahl, Tierwelt Dtl. 18: 166, Fig. 22_{27} (revision of ciliates).

1943 ***Spathidium amphoriforme* Greef-Penard** – Kahl, Infusorien, p. 26, Tafel VI, Fig. 8 (review of ciliates; incorrect spelling of Greeff).

1943 ***Spathidium amphoriforme* var. *rectitoratum* Kahl** – Kahl, Infusorien, p. 27, Tafel VI, Fig. 10 (review of ciliates).

1966 ***Spathidium amphoriforme* Greef** – Dragesco, Arch. Protistenk. 109: 175, Fig. 19 (description of French population from moss; incorrect spelling of Greeff).

1975 ***Spathidium amphoriforme* Greef 1888** – Fryd-Versavel, Iftode & Dragesco, Protistologica 11: 511, 4C, 5C, 6 (description of French population; incorrect spelling of Greeff).

1984 ***Epispathidium amphoriforme* (Greeff, 1888) nov. comb.** – Foissner, Stapfia 12: 82, Abb. 42a–f, Tabelle 21 (description of Austrian population; combination with *Epispathidium*; 2 slides [accession numbers 1981/10, 1984/64] have been deposited in the Biology Centre of the Upper Austrian Museum in Linz [LI]; for details, see nomenclature).

1986 ***Spathidium amphoriforme* (Greef, 1888) Penard, 1922** – Dragesco & Dragesco-Kernéis, Faune tropicale 26: 153, Planche 21E–G (revision of ciliates from Africa; incorrect spelling of Greeff).

1987 ***Epispathidium amphoriforme* var. *rectitoratum* (Kahl, 1930) nov. comb.** – Foissner, Arch. Protistenk. 133: 224 (combination of variety with *Epispathidium*, see nomenclature).

1989 ***Spathidium amphoriforme* (Greeff, 1888)** – Fyda, Acta Protozool. 28: 231, Fig. 1a–e, 2, 3a, Planches I, II, Tableau 1 (description of Polish population, including cell division; site were voucher material deposited not mentioned).

2016 ***Epispathidium amphoriforme* (Greeff, 1888) Foissner, 1984** – Rajter & Vdačný, Zool. Scr. 45: 202 (gene sequence analyses of 2 terrestrial populations from Slovakia based on non-clonal specimens identified in vivo).

2020 ***Epispathidium amphoriforme* (Greeff, 1888) Foissner, 1984** – Kim, Omar & Jung, J. Species Res. 9: 259, Fig. 9A–C (description of Korean population after protargol preparation; two slides with protargol-prepared specimens were deposited at the Nakdonggang National Institute of Biological Resources; accession numbers NNIBRPR11670, NNIBRPR11671).

Nomenclature: The species-group name *amphoriform·is, -is*, *-e* (amphora-shaped) is a composite of the Latin noun *amphora* (vessel, flagon, pitcher, flask, bottle, jar; Brown 1954, p. 86), the thematic vowel *·i-*, and *-form·is*, *-is*, *-e* (Latin adjective [m, f, n]; -shaped; see Hentschel & Wagner 1996, p. 274 at *glómeriformis*), and refers to the jug- or bottle-shaped body (Greeff 1889). There is some uncertainty about the publication year of the work by Greeff. The work is contained in volume 1888 ("Jahrgang 1888") of the journal. However, in the lowest line of the frontpage of the volume the year 1889 appears, indicating that the volume was published just in 1889. Detailed analysis necessary.

The species-group name *rectitoratum* is perhaps a composite of the Latin adjective *rectus, -a, -um* (straight, upright, proper, right; Brown 1954, p. 650), a thematic vowel (?), the Latin noun *ora* (margin, edge, coast; Brown 1954, p. 157), and the suffix *at·us*, *·a*, *·um* (having a feature; Werner 1972, p. 42). Likely it alludes to the fact that the anterior body portion (bearing the oral apparatus) is strongly bent ventrally during swimming (Kahl 1930a, p. 380). Needs more detailed analysis by a linguist.

According to Aescht (2008, p. 142), Foissner (1984) neotypified this species. However, the "neotypification" by Foissner (1984) mentioned in Aescht (2008) is invalid (for details on a valid neotypification, see ICZN 1999, Article 75; see also Aescht 2008, p. 138). A neotypification (which is necessary because the taxonomic status of the present species is uncertain) has to comprise a detailed morphological description including a comprehensive morphometry, preferably also the analysis of the cell division, and molecular analyses. In addition, the neotype has to come as nearly as practicable from the original type locality (ICZN 1999, Article 75.3.6).

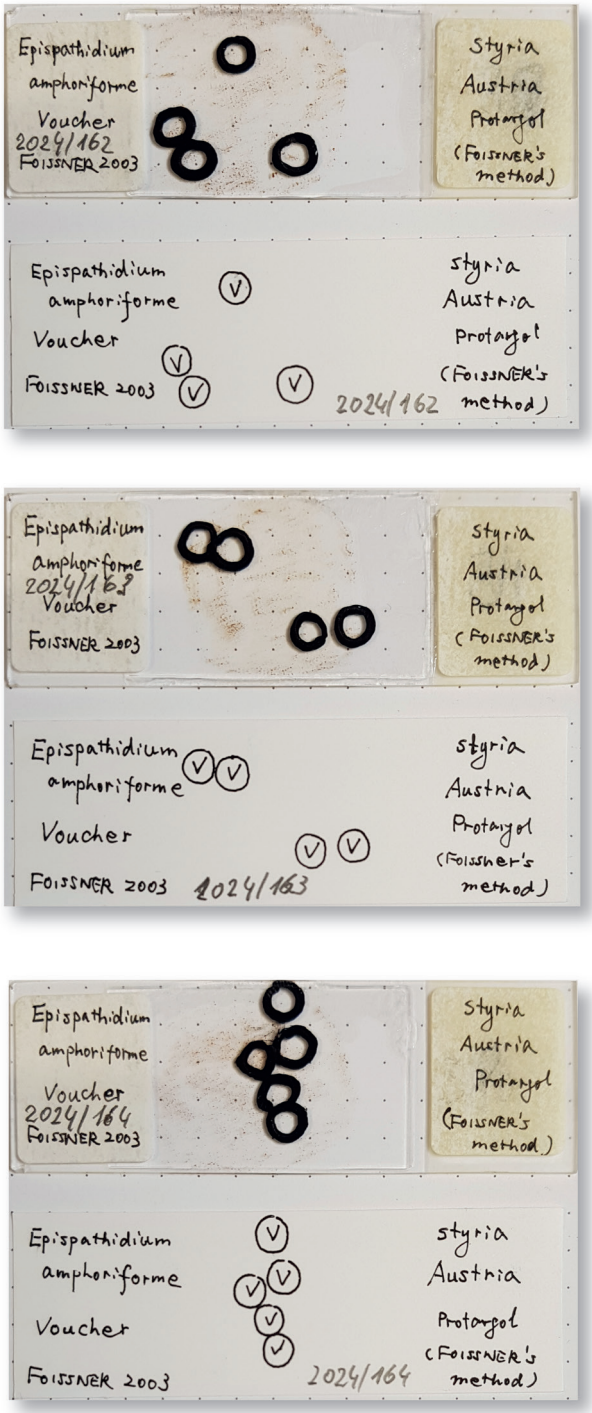

Fig. 6.13a–f *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 (originals. Protargol slides). **a–f:** Slides (a, c, e) and protocols (b, d, f) of voucher specimens (V) of population from Styria, Austria. Accession numbers (LI): 2024/162, 163, 164.

We deposit three voucher slides (Fig. 6.13a–f; accession numbers [LI] 2024/162, 163, 164) of an *Epispathidium amphoriforme* population from Neuwald in Styria (Austria); W. Foissner labeled the slides, but the raw manuscript did not contain data on the morphology (for details on sample site "Neuwald", see Foissner et al. 2005, p. 619).

Brief characterisation according to Foissner (1984): Body size in vivo about 90–150 × 35–65 µm. Body shape distinct amphoriform, oral bulge in badly nourished specimens slightly longer than largest body width, in well-nourished individuals somewhat smaller; in anterior body portion 2–3:1 flatted, behind only slightly to not flattened. Oral bulge distinctly set off from body proper, about 5 µm high, slightly convex, moderately strongly slanted ventrally; in top view narrowly orthogonal and densely filled with about 7 µm long, slightly curved, rod-shaped extrusomes. Macronucleus long,

Table 6.6 Comparison of varieties of *Spathidium amphoriforme* Greeff, 1889 described by Kahl (1930a)

Characteristic	Variety of *Spathidium amphoriforme*		
	amphoriforme[a]	*rectitoratum*[b]	*securiforme*[c]
Body, length	160 µm, little varying	200–240 µm (in older cultures 130–160 µm)	200–300 µm
Body, width	16–25% of body length	about 28% of body length[f]	about 40% of body length[d]
Body, shape	cylindrical	basically as *amphoriforme*, anterior portion abruptly compressed	plump
Somatic kineties, Number	about 20	about 24–28	in large specimens up to 50
Somatic cilia, length	10 µm	as in *amphoriforme*(?)	as in *amphoriforme*(?)
Oral bulge, shape	convex	straight	as in *amphoriforme*
Oral bulge, slope	30°	about 35°[e]	as in *amphoriforme*
Oral bulge, extrusomes	5 µm long	5 µm long, also very numerous in cytoplasm	11–15 µm, fine (in some specimens only 7–8 µm)
Dorsal brush bristles, length	6.0 µm	4.0 µm	2.0–2.5 µm
Nuclear apparatus	Macronucleus very long, constricted, entwined; many micronuclei, adjacent to macronucleus	Macronucleus very long, entwined; micronuclei?[e]	as in *amphoriforme*
Figure	7e in Kahl (1930a)	6.3d in present work	6.3a in present work

[a] Full name when classified in *Spathidium*: *Spathidium amphoriforme amphoriforme* Greeff, 1889. Data from Kahl (1930a, p. 379, diagnosis). Classified as *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 in present work.
[b] Full name when classified in *Spathidium*: *Spathidium amphoriforme rectitoratum* Kahl, 1930. Data from Kahl (1930a, p. 380). Preliminary classified as synonym of *Epispathidium amphoriforme* (Greeff, 1889) Foissner, 1984 in present work.
[c] Full name when classified in *Spathidium*: *Spathidium amphoriforme securiforme* Kahl, 1930. Data from Kahl (1930a, p. 380). Classified as *Epispathidium securiforme* (Kahl, 1930) Foissner, 1984 in present work.
[d] From Fig. 6.3a in present work.
[e] From Fig. 9q in Kahl (1930a).
[f] From Fig. 6.3d in present work.

often strongly tortuous and distinctly nodulated, with many, very small nucleoli. Contractile vacuole terminal, with several excretory pores. Somatic cilia about 13 µm long, dorsal brush bristles about 5 µm long. Between somatic kineties each about six rows of tiny cortical granules. Cytoplasm colourless, with moderately many 3–6 µm sized, globular, yellowish inclusions, many, about 6 µm long spindle-shaped extrusomes and many food vacuoles with

loose content. On average 28 (range 24–38) somatic kineties, longitudinally arranged, anteriorly very densely ciliated, on both sides completely separated from circumoral kinety; those of left side so strongly curved ventrally that they are arranged in parallel to circumoral kinety. Circumoral kinety long orthogonal, composed of basal body pairs from which the nematodesmata originate which extend to mid-body. Dorsal brush shorter than length of oral bulge, kinety 1 and 2 about of same length, kinety 3 only about 40% as long as kineties and 1 and 2.

Remarks: *Spathidium amphoriforme* was originally described relatively detailed, but without an illustration (Greeff 1889). Its taxonomic status is not finally clarified, inter alia, because some important data (e.g., body size, number of somatic kineties) are not known from the original description. There exists a relatively high number of redescriptions (see list above). However, likely not all deal with the same species as indicated, inter alia, by the rather different number of somatic kineties. Kahl (1930a) split *Spathidium amphoriforme* into three varieties, *Spathidium amphoriforme amphoriforme* Greeff, 1889, *Spathidium amphoriforme rectitoratum* Kahl, 1930a, and *Spathidium amphoriforme securiforme* Kahl, 1930a (for note on rank of these "varieties", see Nomenclature at *Epispathidium securiforme*, p. 154; for brief morphological characterisation, see Table 6.6). According to the characterisation by Kahl (1930a; see Table 6.3), at least the populations described by Fryd-Versavel et al. (1975) and Kim et al. (2020) belong to *Epispathidium securiforme* (Kahl, 1930a) Foissner, 1984 because of the high number of somatic kineties (for review, see p. 154). By contrast, *Spathidium amphoriforme rectitoratum* is (preliminary) classified as synonym of *Epispathidium amphoriforme* in the present review (for another opinion, see remarks at *Epispathidium securiforme*).

We suppose that a neotypification (see above) and detailed revision is necessary for a "final" decision about the taxonomic status of *Epispathidium amphoriforme*, including the decision about the generic classification (*Epispathidium* vs. *Spathidium*).

Description: For characterization of population described by Foissner (1984), see above. For description of various populations see individual works mentioned in list of synonyms. However, note that likely not all entries refer to the present species (see remarks).

Resting cyst: Cyst of Type V according to Foissner & Xu (2007, p. 38, Fig. 28V).

Molecular biology: For gene sequence analyses of two populations from Slovakia, see Rajter & Vdačný (2016). Accession numbers of 18S rRNA in GenBank: KT246079 and KT246080.

Occurrence and ecology: Likely a cosmopolitan because recorded from all main biogeographical regions, except for Antarctica (Foissner 1998, p. 203); occurs mainly in terrestrial mosses and leaf litter. The type locality of *Epispathidium amphoriforme* is moss from a wall of the Schlossberg (castle hill; about 50.809°N 08.766°E) of the city of Marburg, Germany (Greeff 1889, p. 131). Penard (1922) found it in mosses from forests and walls from Geneva, Switzerland (see also Kahl 1930b, p. 166). Kahl (1930a, p. 377; 1930b, p. 166) recorded it from terrestrial mosses from Mittenwald, Upper Bavaria, Germany. The type locality of *Spathidium amphoriforme rectitoratum* is the Zillertal (the northern end of this valley is about at 47.40°N 11.83°E; Tyrol, Austria), where Kahl (1930a) found it in (terrestrial) mosses.

Further records substantiated by morphological data: dry mosses and leaf litter, Bavaria, Germany (Wenzel 1953, p. 79); terrestrial? mosses in Villefranche-sur-Mer, France (Dragesco

1966); peat bog in Granvaux near Brinon-sur-Sauldre (47.56°N 02.25°E), France (Fryd-Versavel et al. 1975, p. 511); upper soil layer (0–5cm) of a mixed forest (about 48°22'N 15°34'E, Aescht 2008, p. 142) near the village of Baumgarten, Lower Austria (Foissner 1984, p. 6, 7, for details, see site "FO 16"); forest preservation sites in Eastern Austria, including the sample site "Neuwald" (47°46'N 15°32'E) in Styria (Foissner et al. 2005, p. 627); Yaoundé, Cameroun (Dragesco & Dragesco-Kernéis 1986); mosses from Beskid Niski, Poland (Fyda 1989); leaf litter and terrestrial moss from Slovakia (details see Rajter & Vdačný 2016, p. 202); terrestrial moss from Mt. Yeonhwasan, South Korea (details see Kim et al. 2020, p. 259).

Epispathidium ascendens (Wenzel, 1955) Foissner, 1987

(Fig. 6.14a-r, 6.15a–k)

1955 ***Spathidium ascendens* n. sp.** – Wenzel, Arch. Protistenk. 100: 515, Abb. 1a, b, 2a–j, 3a, b, 4a–c, 5a–c, 6–8, Tabellen 2, 3 (Fig. 6.14a–r; original description; no type material available).

1955 ***Spathidium polymorphum* n. sp.** – Wenzel, Arch. Protistenk. 100: 531, Abb. 10a, b, 11, 12, 14, 15a–j, 16, 17, 18a, b, 19, 20, 21a, b, Tabellen 4, 5 (Fig. 6.15a–k; original description; no type material available; new[?] synonym).

1987 ***Epispathidium ascendens* (Wenzel, 1955) nov. comb.** – Foissner, Sber. öst. Akad. Wiss., Mathematisch-naturwissenschaftliche Klasse, Abt. I 195 (year 1986): 231, Abb. 10a–i, Tabelle 2 (description of an Austrian population, combination with Epispathidium, and neotypification; the neotype slide [accession number 1988/104] has been deposited in the Biology Centre of the Upper Austrian Museum in Linz [LI]; Aescht 2008, p. 144; see nomenclature for details).

2004 ***Epispathidium ascendens*** – Xu & Foissner, J. Euk. Microbiol. 51: 613, Fig. 40–42 (brief description of early stage of conjugation).

2017 ***Spathidium ascendens* Wenzel, 1955** – Jang, Vdačný, Shazib & Shin, J. nat. Hist. 51: 947, Fig. 3a–m, 4a–m, Table 2 (detailed description of Korean population from life and after protargol preparation; locality were voucher material is deposited not mentioned).

Nomenclature: No derivation of the name has been provided in the original description or a later work. The species-group name *ascendens* (from the Latin *ascend*, *ascéndens*) means climb, rise, grow, ascend according to Brown (1954, p. 647) and Hentschel & Wagner (1996, p. 106). We do not know to which feature the species-group name refers. The species-group name *polymorph·us*, *-a*, *-um* ([m, f, n]; New Latin, from Ancient Greek; polymorphous, polymorphic; https://en.wiktionary.org/wiki/polymorphus; accessed 14 Oct 2024) obviously refers to the variable morphology of this population/species.

Aescht (2008, p. 144) wrote that the neotypification is not mentioned in Foissner (1987). This is, however, incorrect because Foissner (1987, p. 234) wrote that the neotype is deposited in the Biology Centre of the Upper Austrian Museum in Linz. However, the question is whether the "neotypification" by Foissner (1987) is valid because the qualifying conditions necessary for a neotypification have not been provided in all details (ICZN 1985, Article 75d; 1999, Article 75.3). For example, Foissner (1987) did not fix the new type locality in the publication because he provided a description composed of data from three populations (Salzburg, Austria; Lower Austria; Greece); according to Aescht (2008, p. 144), the slide of the Salzburg population has been fixed as neotype slide. Jang et al. (2017) obviously did not recognize that Foissner (1987) made a neotypification. The conclusion is that this neotypification needs a detailed analysis to show whether or not it is valid.

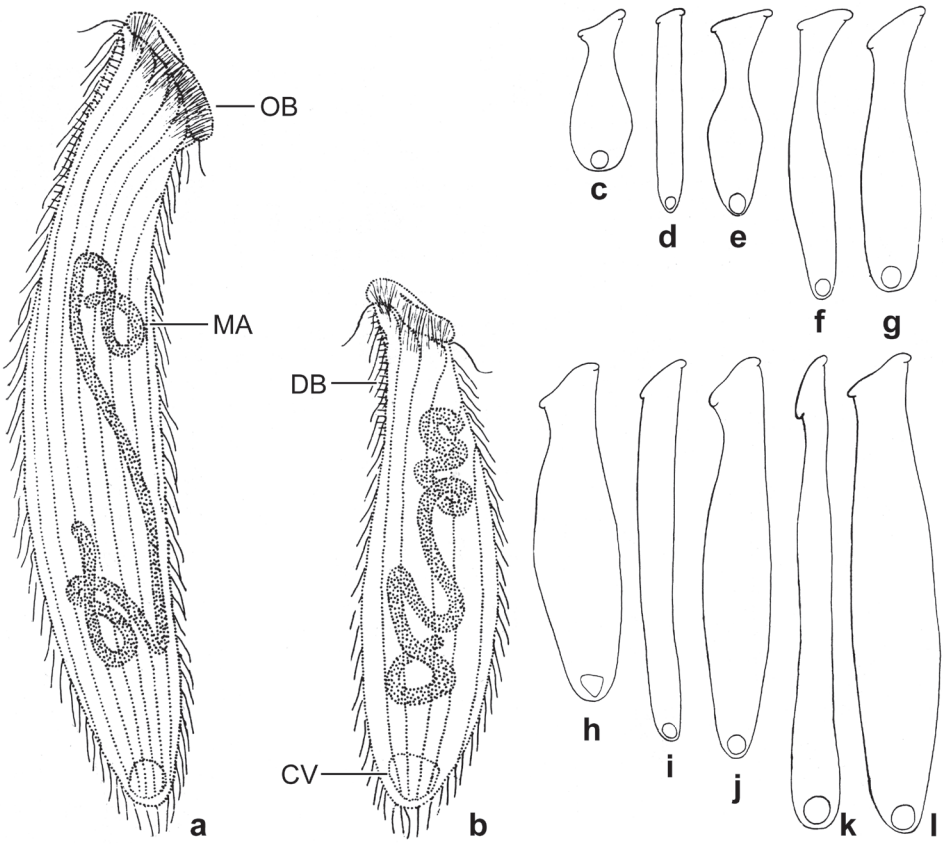

Fig. 6.14a–l *Epispathidium ascendens* (Wenzel, 1955) Foissner, 1987 (from Wenzel 1955. From life?). **a, b:** Right lateral view of two characteristic specimens, ca. 130 µm, ca. 87 µm. No details have been provide in the figure legend about the method. We suppose that the body shape is from life while the ciliature is shown after osmium-toluidin method. **c–l:** Shape variants in lateral view, c = ca. 55 µm (all specimens drawn to scale). CV – contractile vacuole, DB – dorsal brush, MA – macronucleus, OB – oral bulge.

Characterisation of population described by Foissner (1987) (neotype population, see nomenclature; for illustrations, see Fig. 10a–i in Foissner 1987):[19] Body size in vivo 130–200 × 25–50 µm. Body outline typically spathidiforme, anteriorly slightly to distinctly slanted, posteriorly tapered and narrowly rounded. Neck region about 2:1 flattened, posteriorly round in cross-section. Oral bulge about 5 µm high, wider than the neck-region, in middle portion slightly to distinctly recessed, ventrally and dorsally slightly opened and widened, in top view thus dumbbell-shaped; packed with 8–12 µm long, rod-shaped extrusomes, many of them also present in cytoplasm. Macronucleus long ribbon-like, in the Austrian populations only rarely (likely accidentally), in the Greece population usually with a straight, middle portion. Micronuclei lacking (or overlooked? See Table 2 in Foissner et al. 1987 and comment below). Contractile vacuole terminal; many excretory pores in the

[19] The in vivo observations are based on three populations from various localities (details, see occurrence and ecology).

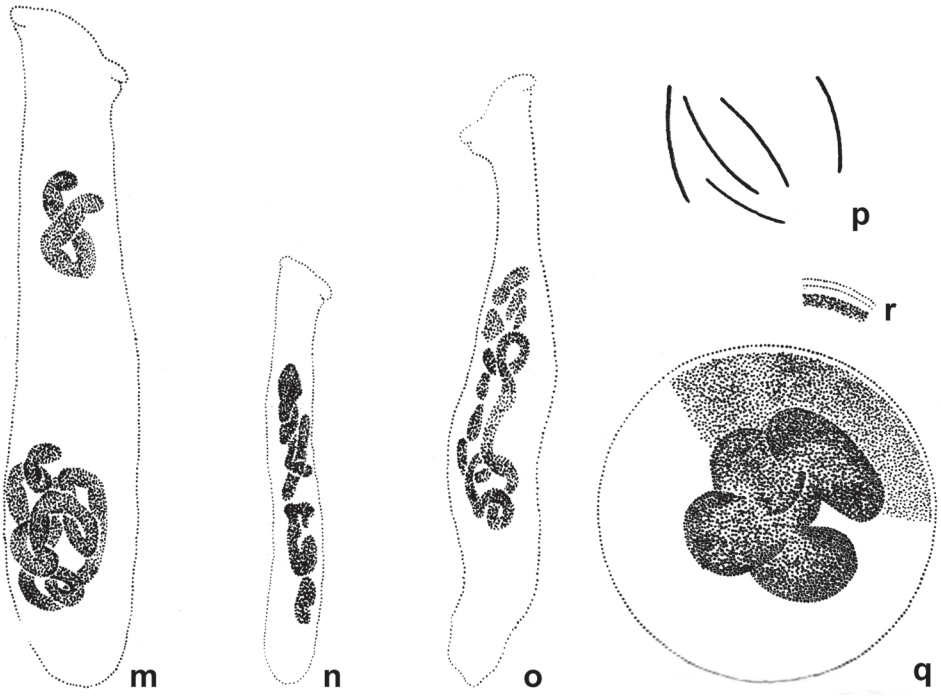

Fig. 6.14m–r *Epispathidium ascendens* (Wenzel, 1944) Foissner, 1987 (from Wenzel 1955. m, Feulgen-staining; n, o, gallocyanin; p, after stimulation with formol, darkfield; q, r, from life). **m–o:** Specimens with partly fragmented macronucleus, ca. 115 µm, ca. 73 µm, ca. 103 µm. **p:** Ejected extrusomes, ca 10 µm long. **q:** Resting cyst, ca. 40 µm. **r:** Cyst wall, about 3 µm thick.

centre of the posterior pole. Somatic cilia about 10 µm long, bristles of dorsal brush about 2 µm long. Several rows of small, colourless granules (mucocysts) between each two somatic kineties. Cytoplasm colourless, usually hyaline, with few globular inclusions and 2–5 µm-sized, vacuoles which appear to be empty. Movement moderately rapid gliding or slowly rotating about main body axis.

On average 21 (range 20–25) moderately densely ciliated somatic kineties, usually distinctly set off from the circumoral kinety which is composed of dikinetids; somatic kineties anteriorly only moderately densely ciliated; kineties on left side not always distinctly curved ventrally. Kineties of dorsal brush almost of equal length, middle one usually longest. Cytopharynx strong.

Remarks: The populations described by Wenzel (1955), Foissner (1987), and Jang et al. (2017) agree in all main features indicating that they are conspecific. Foissner (1987, p. 234) discussed that the classification in *Epispathidium* is not always obvious because the somatic kineties on the left side are not distinctly curved ventrally in each specimen. Jang et al. (2017) preferred a classification in *Spathidium* because the oral ciliature is more reminiscent to *Spathidium* than to *Epispathidium* and, in addition, because it clusters with other *Spathidium* species (e.g., *Spathidium stammeri* Wenzel, 1959; *Spathidium* sp.; *Spathidium*

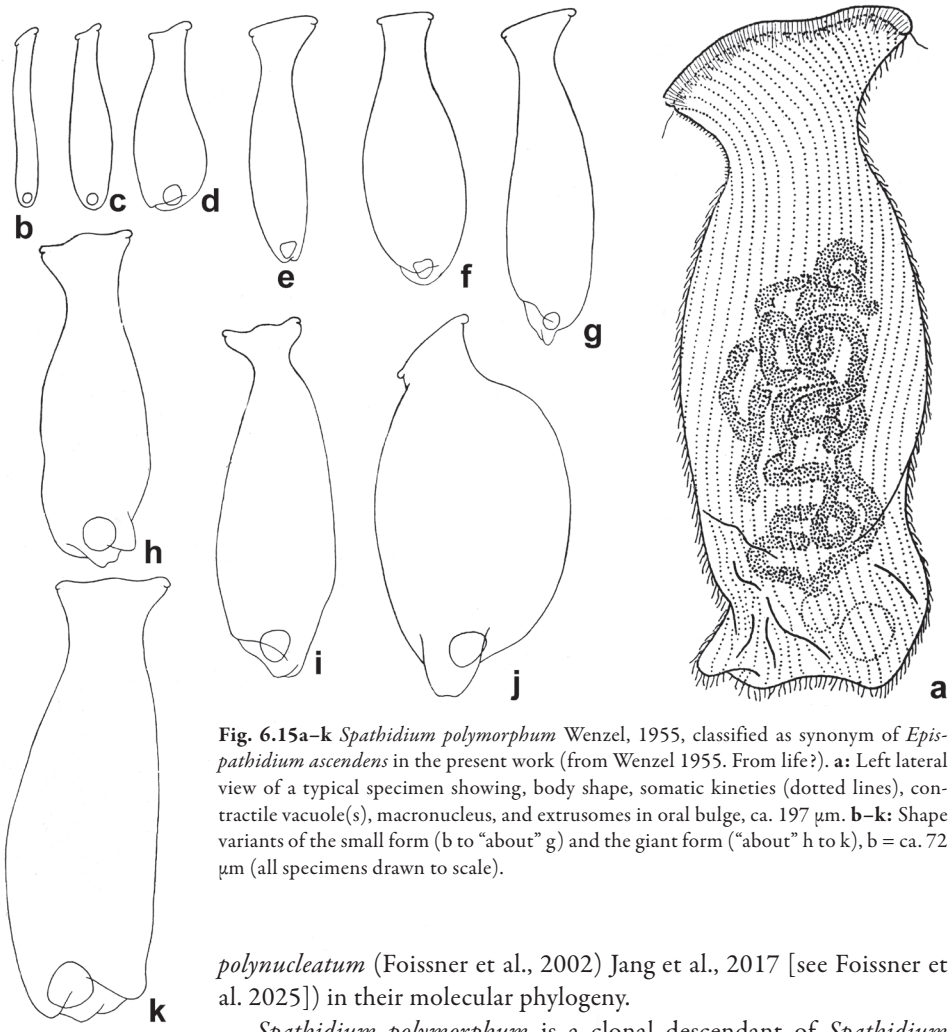

Fig. 6.15a–k *Spathidium polymorphum* Wenzel, 1955, classified as synonym of *Epispathidium ascendens* in the present work (from Wenzel 1955. From life?). **a:** Left lateral view of a typical specimen showing, body shape, somatic kineties (dotted lines), contractile vacuole(s), macronucleus, and extrusomes in oral bulge, ca. 197 µm. **b–k:** Shape variants of the small form (b to "about" g) and the giant form ("about" h to k), b = ca. 72 µm (all specimens drawn to scale).

polynucleatum (Foissner et al., 2002) Jang et al., 2017 [see Foissner et al. 2025]) in their molecular phylogeny.

Spathidium polymorphum is a clonal descendant of *Spathidium ascendens*. It differs very strongly from *Spathidium ascendens* in some morphological and physiological features causing Wenzel (1955) to establish a new species for this giant form. We preliminary classify *Spathidium polymorphum* as objective synonym of *Epispathidium ascendens* because all these cells are from the same clone. More detailed studies are needed to support one of these two hypotheses (one species vs. already two species).

Jang et al. (2017) reported that the micronuclei are near or attached to the macronucleus at varying positions; the exact number was difficult to determine due to similar-sized cytoplasmic globules nearby. By contrast, the clone studied by Wenzel (1955) was amicronucleate according to analyses of several hundred specimens stained by various methods (Wenzel 1955, p. 521). Foissner (1987) made no specific comment on this feature, that is, he did not mention this organelle in the text, and from his "Tabelle 2" it is unclear if he checked this feature at all.

Description: For detailed description of German populations, see Wenzel (1955, p. 518, 531; in German); for brief morphological characterisation of both *Epispathidium ascendens* and its supposed synonym *Spathidium polymorphum*, see next paragraph. For characterization of neotype population, see above (for problems on neotypification, see Nomenclature). For detailed description of Korean population, see Jang et al. (2017, p. 947).

Epispathidium ascendens (from Wenzel 1955; Fig. 6.14a–r; for more figures and more data, see Wenzel 1955): Body size rather variable, on average 135 × 36 µm (n = 342; method not indicated). Body outline highly variable (Fig. 6.14a–l). Macronucleus long band-shaped (Fig. 6.14a, b); in some cases, fragmented (Fig. 6.14m–o). Clone analysed without micronucleus. 8–14 somatic kineties per side, rarely only five (n = 72; Fig. 6.14a, b). Contractile vacuole in rear body portion, during diastole originating from several vacuoles. Cytoplasm of starving cells translucent and finely granulated. Extrusomes very fine, difficult to recognize when not ejected; on average 8.5 µm (range 6–11 µm, n = 86) long according to Tabelle 2 in Wenzel (1955; state [resting or ejected] not clearly indicated); ejected extrusomes very fine and slightly curved (Fig. 6.14p). Somatic cilia about 6.5 µm long (n = 40; method [in vivo or after fixation] not indicated); cilia of circumoral kinety about 9.5 µm long (n = 40); dorsal bristles about 1.5 µm long (n = 20); dorsal brush 20–35 µm long (n = 25); width of oral bulge ("Lippenbreite") 1.6–3.0 µm (n = 35).

Spathidium polymorphum (from Wenzel 1955; Fig. 6.15a–k; for more figures and more data, see Wenzel 1955): Body size highly variable (Fig. 6.15a–k; n = 680; see Tabelle 4 in Wenzel 1955); body length ranges from size-class 40–70 µm to class 281–310 µm, most specimens within class 161–190 µm; body width 10–25 µm to 101–103 µm, most within class 41–55 µm. Body outline highly variable (Fig. 6.15a–k). Macronucleus band-shaped, often thicker, and always longer and stronger than in *Epispathidium ascendens* (Fig. 6.15a). Contractile vacuole, extrusomes, details of ciliature (length of cilia, etc.) as in *Epispathidium ascendens* (for details, see Tabelle 5 in Wenzel 1955). 9–30 somatic kineties per side (n = 74). Cytoplasm (except in neck region) opaque due to glycogen globules.

Resting cysts: Resting cyst of *Epispathidium ascendens* 30–44 µm, on average 38 µm (n = 31) in diameter; wall about 3 µm thick, smooth, clear translucent (Fig. 6.14q, r; Wenzel 1955). This agrees with the detailed description by Jang et al. (2017, p. 949) who characterize the cysts as colourless, about 25–40 µm across, and with a smooth wall.

Resting cyst of synonym *Spathidium polymorphum* 30–88 µm (on average 65 µm) in diameter (n = 48; Wenzel 1955, p. 535).

Cell division and budding: In *Spathidium polymorphum*, Wenzel (1955, p. 525) reported ordinary cell division, but also budding (e.g., his Abb. 10a, b, 11, 12, 17). For details, see original description.

Conjugation: Wenzel (1955, his Abb. 8) found five conjugating pairs (for details, see Wenzel 1955, p. 524). For the ciliary pattern in the oral region of three early conjugating pairs, see Xu & Foissner (2004). For some information on conjugation of the

Molecular biology: For gene sequence analyses of the Korean population, see Jang et al. (2017, p. 941, 969). Accession number of 18S rRNA in GenBank: KY556643.

The tree based on 18S rRNA shows that *Spathidium* is likely a non-monophyletic group. Besides many *Spathidium* species, the cluster contains members of the genera *Arcuospathidium*, *Teuthophrys*, *Lagynophrya*, *Enchelys*, *Balantidion*, *Apobryophyllum*, *Cultellothrix*,

Trachelophyllum, *Epispathidium*, *Enchelyodon*, *Pseudoholophrya*, *Acaryophrya*, and *Semispathidium* (Jang et al. 2017, their Fig. 16). This demonstrates that we are just at the beginning of understanding the phylogeny of this group of ciliates.

Occurrence and ecology: According to Foissner (1998, p. 203), *Epispathidium ascendens* is a true cosmopolitan species because recorded from all main biogeographic regions. Wenzel (1955) discovered it in moos lawn collected from the Botanical Gardens of the Botanical Institute of the University of Erlangen, Bavaria, Germany; the sample was collected on 10 Jul 1953 (Wenzel 1955, p. 516, footnote 1). All cultures studied by Wenzel (1955) are clones of a single specimen of *Spathidium ascendens* (Wenzel 1955, p. 516). Wenzel (1955, p. 516) used a 6–14 days-old suspension of yolk in tap water. To this medium a *Dexiostoma campylum* clone was added serving as food for *Epispathidium ascendens*.

The in vivo observations of Foissner (1987) are based on three populations (Seekirchen, Austria; Tullnerfeld, Austria; Peloponnese, Greece); however, the description of the ciliature and the morphometry is based on protargol preparations from the population from Seekirchen; thus, this area is the new type locality (see, however, nomenclature for problems with the neotypification): upper soil layer (0–10 cm) of a conventionally farmed field (pH 5.7; 600 m above sea-level) near the village of Seekirchen am Wallersee (47.89°N 13.12°E), Salzburg, Austria (Foissner 1987, p. 217, 218; see also experimental area "Versuchsfläche H" in Foissner et al. 1987, p. 334). The Korean population was isolated from leaf litter and soil from Daewangam Park (35.494°N 129.443°E), Ilsan-dong, Dong-gu, Ulsan (Jang et al. 2017, p. 941).

Epispathidium ascendens feeds on ciliates (*Colpoda fastigata*; Foissner 1987, p. 234). Biomass of 10^6 specimens about 64 mg (Foissner 1987b, p. 123; 1998, p. 203).

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Note on *Epispathidium papilliferum*: According to Weiss et al. (2022), *Legendrea loyezea* Fauré-Fremiet, 1908, type species of *Legendrea* Fauré-Fremiet, 1908, forms a cluster with *Epispathidium* sp. + *Epispathidium papilliferum*, however, with low support.

Note on *Epispathidium amphoriforme* from Neuwald (see p. 199): A description of this population will be provided in a later work.

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Systematic index

The index contains all ciliate names mentioned in the book, including vernacular names for example, haptorids. Designations as, for example, "haptorid ciliates" are mentioned under the corresponding vernacular name, that is, "haptorids" in present example. Names in singular (e.g., haptorid) are mentioned under the plural version (e.g., haptorids). The index is two-sided, that is, species appear both with the genus-group name first (for example, *Apospathidium atypicum*) and with the species-group name first (*atypicum*, *Apospathidium*). Valid (mainly in W. Foissner's judgement) species and genera treated in detail are in boldface italics print. Valid taxa not treated in detail in the present book, invalid taxa, junior homonyms, synonyms, outdated combinations, incorrect spellings, and nomina nuda are not in bold. Suprageneric taxa are represented in normal type, valid ones treated in detail in the present work in boldface. A boldface page number indicates the beginning of the description of a valid taxon. "T" indicates the location of the table with the morphometric characterisation; "K" marks a key (e.g., of the genus *Apospathidium*) and the page where a taxon is mentioned in a key. The names on the slide figures and the names of the subchapter "Summary of nomenclatural acts and taxa described in Chapters 1–13" (see Chapter 1, pp. 18–20) are not included.

Table index