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# Ontogenesis of *Dileptus terrenus* and *Pseudomonilicaryon brachyproboscis* (Ciliophora, Haptoria)

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ABSTRACT. Dileptids are haptorid ciliates with a conspicuous proboscis belonging to the oral apparatus and carrying a complex, unique ciliary pattern. We studied development of body shape, ciliary pattern, and nuclear apparatus during and after binary fission of *Dileptus terrenus* using protargol impregnation. Additional data were obtained from a related species, *Pseudomonilicaryon brachyproboscis*. Division is homothetogenic and occurs in freely motile condition. The macronucleus is homomeric and condenses to a globular mass in mid-dividers. The proboscis appears in late mid-dividers as a small convexity in the opisthe's dorsal brush area and maturates post-divisionally. The oral and dorsal brush structures develop by three rounds of basal body proliferation. The first round generates minute anarchic fields that will become circumoral kinetofragments, while the second round produces the perioral kinety on the right and the preoral kineties on the dorsal opisthe's side. The dorsal brush is formed later by a third round of basal body production. The formation of various *Spathidium*-like body shapes and ciliary patterns during ontogenesis and conjugation of *Dileptus* shows a close relationship between spathidiids and dileptids. On the other hand, the peculiarities of the dileptid morphology and ontogenesis indicate a long, independent evolution.

**Key Words.** Dorsal brush, evolution, soil ciliates, spathidiids, stomatogenesis.

HAPTORIDS are a highly diverse group of ciliates characterized by toxicysts, a dorsal brush, and a telokinetal stomatogenesis (Corliss 1979; Foissner 1996; Foissner and Foissner 1988). Within the order Haptorida, dileptids represent a special cytoarchitectural type due to the bi-or tripartition of the body into a proboscis, trunk, and tail (Dragesco 1963; Kahl 1931). Further, the dileptid oral infraciliature is much more complex than in other haptorids: (1) the circumoral kinety is composed of dikinetids in the proboscis, while that of oralized somatic monokinetids around the oral bulge opening, which is situated at the base of the proboscis; (2) the oral dikinetids do not bear nematodesmata, unlike the non-ciliated oralized somatic monokinetids; and (3) the right branch of the circumoral kinety is accompanied by a perioral kinety, while the left branch is associated with many short, oblique preoral kineties (Foissner 1997; Foissner and Foissner 1988; Golińska 1991; Grain and Golińska 1969; Kink 1976).

The genus *Dileptus* plays an import role in understanding haptorid evolution because it branches basally in molecular phylogenies (Strüder-Kypke et al. 2006). Further, its oral apparatus seems to be a composite of special features found in related taxa: it has oralized somatic monokinetids, which typically occur in the Enchelyina and it has oral dikinetids, which typically occur in the Spathidiida (Foissner and Foissner 1988; Foissner and Xu 2007; Golińska 1995). Only one attempt has been made to explain the relationship between dileptids and other haptorids: Xu and Foissner (2005) speculated that spathidiids evolved from a *Dileptus*-like ancestor by reduction of the proboscis. This hypothesis was later supported by the formation of a *Spathidium*-like body shape and ciliary pattern during conjugation and post-conjugational reorganization of *Dileptus* (Vd'ačný and Foissner 2008a). The present investigation adds a further argument: *Protospathidium*- and *Epispathidium*-like body shapes and ciliary patterns are transiently formed during ontogenesis.

Data on binary fission of *Dileptus* have been reported for *Dileptus anatinus*, *Dileptus cygnus*, *Dileptus margaritifer*, and *Dileptus visscheri* (Bohatier and Kink 1977; Golińska 1972, 1995), as well as for three *Paradileptus* species: *Paradileptus conicus*, *Paradileptus elephantinus*, and *Paradileptus ovalis* (Fryd-Versavel, Iftode, and Dragesco 1975; Huber-Pestalozzi 1945). However, most of these studies are very incomplete, providing only a single stage and/or a few schematic figures or micrographs. The exceptions are the detailed investigations of Golińska (1972, 1995) who studied mainly the formation of the opisthe's infraciliature, using transmission electron microscopy and protargol impregnation. Nevertheless, the knowledge on the development of body shape, ciliary pattern, and nuclear apparatus during and after binary fission of *Dileptus* is still insufficient. Therefore, we studied ontogenesis in *Dileptus terrenus* and *Pseudomonilicaryon brachyproboscis*, using silver impregnation and showing the process by detailed ink drawings. This demonstrates that ontogenesis of *Dileptus* sensu lato is much more complex than in other haptorids, reaching an extent comparable to that found in ''higher'' ciliates.

## MATERIALS AND METHODS

*Dileptus terrenus* Foissner 1981 was found in an up to 10-cm-thick accumulation of washed-up plant debris, including filamentous algae, macrophytes, duckweeds, and some soil, pH 6, at the bank of the Titicaca Lake, near the town of Puno, Peru (15°22′S 69°30′W). This material was air-dried and then used to set up a non-flooded Petri dish culture, as described by Foissner, Agatha, and Berger (2002). A semi-pure culture of *D. terrenus* was established with some drops of the percolate from the non-flooded Petri dish culture and Eau de Volvic enriched with some crushed wheat grains to stimulate growth of bacteria and prey protozoa. Additional data were obtained from another dileptid ciliate, *P. brachyproboscis* Vd'ačný and Foissner 2008. The species occurred in a non-flooded Petri dish culture set up with terrestrial mosses and *Pinus* needles from the Peloponnese, Greece (38°01′N 21°57′E). Unfortunately, late dividers were not found in the protargol slides.

Growing cultures were fixed in toto with Stieve's solution and impregnated with protargol protocol A described in Foissner (1991). The ontogenetic events were reconstructed from these preparations, which show concomitantly body shape, ciliary pattern, and nuclear apparatus. Figures were made with a drawing device. Counts and measurements were performed at a magnification of 1,000X. The preparations were deposited in the Biologiezentrum of the Oberösterreichische Landesmusem in Linz (LI), Austria. Relevant specimens were marked by blank ink circles on the coverslip.

Interphase terminology is based on Corliss (1979). Ontogenesis terminology is according to Foissner (1996). For designating ventral, lateral, and dorsal ciliary rows, see Fig. 54. Division stages

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are distinguished as follows: very early dividers are characterized by the proliferation of basal bodies in the dorsal kineties slightly posterior to the mid-body, while early dividers have developed oral kinetofragments. Mid-dividers have a continuous circumoral kinety and the macronucleus commences to condense. Late dividers have a dumbbell-shaped macronucleus, while the macronucleus is divided into two pieces connected by a fiber-like structure in very late dividers.

## RESULTS

**Morphology of *Dileptus terrenus* (Fig. 1–4; Table 1).** The morphostatic cells match well the description of *D. terrenus* (Foissner 1981, 1984). Thus, mainly features relevant to the present study will be briefly described. Body size is about 280 × 50 μm with a length:width ratio of an average of 5.9:1 in protargol preparations and with the proboscis occupying about 33% of body length (Table 1). The macronucleus is cylindroidal and more or less curved, typically accompanied by a single micronucleus about 4 μm in diameter. A row of contractile vacuoles occurs on the dorsal side of the trunk and the proximal half of the proboscis (Fig. 4). There is only one type of rod-shaped extrusomes, 4 μm long and anchored to the oral bulge of the proboscis. The cilia are arranged in an average of 27 longitudinal rows, which gradually shorten anteriorly to produce a suture along the right side of the oral bulge (Fig. 4, arrows), except for the perioral kinety, which extends with densely spaced basal bodies to tip of proboscis. The left side of the proboscis has a conspicuous blank stripe because most left side ciliary rows are shortened at the level of the oral bulge opening (Fig. 2, 3, asterisks). The multi-rowed, staggered dorsal brush extends on the dorsal surface of the proboscis (Fig. 1). The structure of the oral apparatus is as in other dileptids: (1) the oral bulge opening is located at the base of the proboscis; (2) there are an internal and external, conical oral basket; (3) the circumoral kinety is dikinetidal, except for the monokinetidal portion around the oral bulge opening; and (4) the preoral kineties are composed of two to three narrowly to ordinarily spaced monokinetids distinctly inclined to the left branch of the circumoral kinety (Fig. 2–4).

**Ontogenesis of *Dileptus terrenus* (Fig. 5–34; Table 1).** Many dividers and post-dividers were found in the protargol slides. Thus, each stage was observed in at least four specimens, and the quantitative features could be underpinned by statistics. We could not follow the origin of the contractile vacuole pattern because the excretory pores impregnated too faintly.

*Division mode.* Fission is homothetogenic, occuring in freely motile (non-encysted) condition. Stomatogenesis is holotelokinetal. The parental oral apparatus and dorsal brush are not reorganized.

*Body changes and development of proboscis.* Very early dividers are longer than morphostatic specimens by an average of about 30 μm, while body width and the ratio of body and proboscis length hardly change (~31% vs. 34%, Table 1). Thus, early dividers are the largest and most slender cells because they are longer (322 vs. 278 μm), but not significantly wider than morphostatic specimens (50 vs. 48 μm; Fig. 16). In contrast to spathidiids, there is neither a slight indentation in the prospective fission area nor division blebs. In mid-dividers, when the macronucleus condenses, the body shortens and broadens from 322 × 50 to 303 × 56 μm: these cells are the smallest and stoutest dividers (Fig. 17, 18). At this stage, a minute bare protuberance, the precursor of the oral bulge, develops along the prospective anterior end of the opisthe, dividing the cell into a conical posterior daughter shorter by about one-fifth than the broad proter (Fig. 11, 12, 17, 18). In late mid-dividers, a remarkable process commences: the proboscis bud develops as a small convexity in the opisthe's brush area underneath the developing division furrow (Fig. 19, arrow). In late

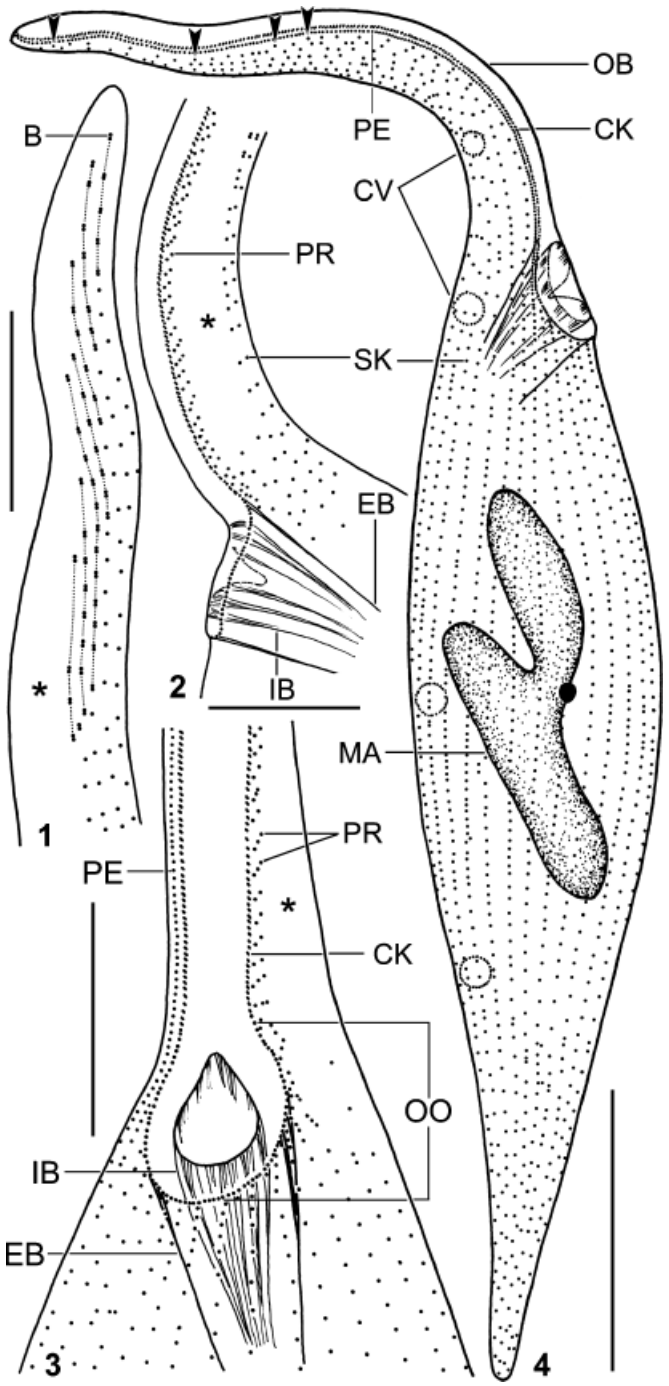


Fig. **1–4.** *Dileptus terrenus*, ciliary pattern of morphostatic specimens from a Peruvian population after protargol impregnation. **1.** Dorsal view of proboscis, showing the staggered dorsal brush rows (connected by minute dots). **2.** Left side ciliary pattern of proximal proboscis area. **3.** Infraciliature in oral region. **4.** Ciliary pattern of right side and nuclear apparatus of a representative specimen, length 318 μm. Arrowheads mark gradually shortened somatic kineties in anterior right portion of proboscis. Asterisks denote barren stripe on left side of proboscis. B, dorsal brush; CK, circumoral kinety; CV, contractile vacuoles; EB, external basket; IB, internal basket; OB, oral bulge; OO, oral bulge opening; PE, perioral kinety; PR, preoral kineties; MA, macronucleus; SK, somatic kinety. Scale bars 20 μm (Fig. 1–3) and 50 μm (Fig. 4).

Table **1.** Morphometric data on morphostatic cells, dividers, and post-dividers of *Dileptus terrenus*.

| Characteristics[a] | Stage[b] | Mean | M | SD | SE | CV | Min | Max | *n* |
|---|---|---|---|---|---|---|---|---|---|
| Body, length | Morphostatic | 278.4 | 283.0 | 23.1 | 5.0 | 8.3 | 227.0 | 315.0 | 21 |
| | Very early divider | 310.9 | 306.0 | 30.6 | 6.7 | 9.8 | 265.0 | 372.0 | 21 |
| | Early divider | 322.1 | 323.0 | 32.3 | 7.0 | 10.0 | 252.0 | 369.0 | 21 |
| | Mid-divider | 303.5 | 303.0 | 25.6 | 5.6 | 8.4 | 246.0 | 350.0 | 21 |
| | Late divider | 308.3 | 313.0 | 32.5 | 13.3 | 10.6 | 249.0 | 340.0 | 6 |
| | Very late divider | 310.3 | 314.0 | — | — | — | 268.0 | 345.0 | 4 |
| | Proter post-divider | 207.6 | 211.0 | 21.0 | 4.6 | 10.1 | 153.0 | 240.0 | 21 |
| | Opisthe post-divider | 169.7 | 172.0 | 25.4 | 5.5 | 15.0 | 137.0 | 223.0 | 21 |
| Body, width | Morphostatic | 47.8 | 49.0 | 5.8 | 1.3 | 12.2 | 35.0 | 56.0 | 21 |
| | Very early divider | 49.7 | 49.0 | 4.5 | 1.0 | 9.1 | 44.0 | 60.0 | 21 |
| | Early divider | 50.1 | 51.0 | 4.0 | 0.9 | 8.0 | 43.0 | 63.0 | 22 |
| | Mid-divider | 55.8 | 56.0 | 5.7 | 1.3 | 10.3 | 43.0 | 70.0 | 21 |
| | Late divider | 52.7 | 54.0 | 3.3 | 1.3 | 6.2 | 49.0 | 56.0 | 6 |
| | Very late divider | 59.4 | 61.0 | — | — | — | 48.0 | 69.0 | 4 |
| | Proter post-divider | 48.3 | 49.0 | 5.7 | 1.3 | 11.9 | 40.0 | 63.0 | 21 |
| | Opisthe post-divider | 43.5 | 43.0 | 4.4 | 0.9 | 10.0 | 36.0 | 53.0 | 21 |
| Anterior body end to oral opening, distance | Morphostatic | 94.0 | 94.0 | 12.3 | 2.7 | 13.1 | 78.0 | 113.0 | 21 |
| | Very early divider | 97.1 | 102.0 | 12.6 | 2.8 | 13.0 | 70.0 | 114.0 | 21 |
| | Early divider | 101.7 | 102.0 | 11.5 | 2.5 | 11.3 | 72.0 | 125.0 | 21 |
| | Mid-divider | 92.3 | 93.0 | 13.1 | 2.9 | 14.2 | 59.0 | 117.0 | 21 |
| | Late divider | 81.4 | 80.0 | 10.7 | 4.4 | 13.1 | 70.0 | 100.0 | 6 |
| | Very late divider | 88.8 | 89.0 | — | — | — | 74.0 | 103.0 | 4 |
| | Proter post-divider | 95.4 | 98.0 | 13.1 | 2.9 | 13.7 | 62.0 | 113.0 | 21 |
| | Opisthe post-divider | 50.7 | 51.0 | 10.2 | 2.2 | 20.1 | 30.0 | 67.0 | 21 |
| Proter, length | Very early divider | 177.8 | 176.0 | 19.4 | 4.2 | 10.9 | 144.0 | 220.0 | 21 |
| | Early divider | 180.5 | 180.0 | 19.3 | 4.2 | 10.7 | 142.0 | 215.0 | 21 |
| | Mid-divider | 169.5 | 168.0 | 17.6 | 3.8 | 10.4 | 145.0 | 205.0 | 21 |
| | Late divider | 166.5 | 172.0 | 17.9 | 7.3 | 10.7 | 142.0 | 191.0 | 6 |
| | Very late divider | 174.5 | 176.0 | — | — | — | 149.0 | 198.0 | 4 |
| Proter, width | Very early divider | 49.5 | 49.0 | 4.7 | 1.0 | 9.4 | 42.0 | 60.0 | 21 |
| | Early divider | 50.1 | 51.0 | 4.0 | 0.9 | 8.0 | 43.0 | 63.0 | 22 |
| | Mid-divider | 55.8 | 56.0 | 5.7 | 1.3 | 10.3 | 43.0 | 70.0 | 21 |
| | Late divider | 52.7 | 55.0 | 3.3 | 1.3 | 6.2 | 49.0 | 56.0 | 6 |
| | Very late divider | 59.4 | 61.0 | — | — | — | 48.0 | 69.0 | 4 |
| Proter, oral opening to proter end, distance | Very early divider | 80.7 | 78.0 | 10.6 | 2.3 | 13.1 | 69.0 | 106.0 | 21 |
| | Early divider | 78.8 | 84.0 | 11.6 | 2.5 | 14.7 | 55.0 | 94.0 | 21 |
| | Mid-divider | 77.2 | 75.0 | 9.9 | 2.2 | 12.8 | 67.0 | 98.0 | 21 |
| | Late divider | 85.1 | 88.0 | 13.9 | 5.7 | 16.3 | 61.0 | 100.0 | 6 |
| | Very late divider | 85.8 | 87.0 | — | — | — | 68.0 | 101.0 | 4 |
| Opisthe, length | Very early divider | 133.1 | 134.0 | 13.5 | 2.9 | 10.1 | 110.0 | 152.0 | 21 |
| | Early divider | 142.2 | 141.0 | 16.4 | 3.5 | 11.6 | 110.0 | 171.0 | 22 |
| | Mid-divider | 134.0 | 135.0 | 12.3 | 2.7 | 9.2 | 102.0 | 150.0 | 21 |
| | Late divider | 135.7 | 140.0 | 9.1 | 3.7 | 6.7 | 121.0 | 144.0 | 6 |
| | Very late divider | 147.4 | 146.0 | — | — | — | 137.0 | 162.0 | 4 |
| Opisthe, width | Very early divider | 46.1 | 46.0 | 4.3 | 0.9 | 9.4 | 38.0 | 59.0 | 21 |
| | Early divider | 45.3 | 46.0 | 4.9 | 1.0 | 10.8 | 34.0 | 58.0 | 22 |
| | Mid-divider | 50.3 | 51.0 | 7.4 | 1.6 | 14.7 | 34.0 | 69.0 | 21 |
| | Late divider | 47.0 | 47.0 | 3.2 | 1.3 | 6.7 | 42.0 | 52.0 | 6 |
| | Very late divider | 53.1 | 54.0 | — | — | — | 42.0 | 63.0 | 4 |
| Macronucleus, total length | Morphostatic | 81.5 | 80.0 | 17.1 | 3.7 | 21.0 | 62.0 | 128.0 | 21 |
| | Very early divider | 84.2 | 86.0 | 12.3 | 2.7 | 14.7 | 59.0 | 105.0 | 21 |
| | Early divider | 99.7 | 93.0 | 16.3 | 3.5 | 16.3 | 78.0 | 146.0 | 22 |
| | Mid-divider | 47.8 | 47.0 | 9.2 | 2.0 | 19.3 | 31.0 | 64.0 | 21 |
| | Late divider | 103.5 | 101.0 | 10.6 | 4.3 | 10.3 | 93.0 | 120.0 | 6 |
| | Very late divider | 100.4 | 100.0 | — | — | — | 87.0 | 116.0 | 4 |
| | Proter post-divider | 82.3 | 82.0 | 19.3 | 4.2 | 23.5 | 43.0 | 121.0 | 21 |
| | Opiste post-divider | 70.8 | 65.0 | 26.7 | 5.8 | 37.7 | 30.0 | 124.0 | 21 |
| Micronucleus, largest diameter | Morphostatic | 3.6 | 4.0 | — | — | — | 3.0 | 4.0 | 14 |
| | Very early divider | 4.4 | 4.0 | 0.8 | 0.3 | 19.0 | 3.0 | 6.0 | 9 |
| | Early divider | 6.1 | 6.0 | 1.0 | 0.3 | 16.4 | 4.0 | 7.0 | 12 |
| | Mid-divider | 6.3 | 6.0 | 1.6 | 0.6 | 25.4 | 5.0 | 10.0 | 8 |
| | Late divider | 4.0 | 4.0 | 0.0 | 0.0 | 0.0 | 4.0 | 4.0 | 4 |
| | Very late divider | 4.0 | 4.0 | — | — | — | 4.0 | 4.0 | 3 |
| | Proter post-divider | 4.0 | 4.0 | 0.5 | 0.2 | 12.5 | 3.0 | 5.0 | 9 |
| | Opisthe post-divider | 3.9 | 4.0 | 0.9 | 0.3 | 22.3 | 3.0 | 6.0 | 12 |
| Ciliary rows, number | Morphostatic | 26.9 | 27.0 | 2.0 | 0.4 | 7.4 | 24.0 | 31.0 | 21 |
| Preoral kineties, number | Morphostatic | 47.1 | 46.0 | 5.1 | 1.1 | 10.9 | 41.0 | 58.0 | 21 |
| Dorsal brush dikinetids, total number | Morphostatic | 55.3 | 56.0 | 6.6 | 1.4 | 12.0 | 43.0 | 69.0 | 21 |

[a]Data based on mounted, protargol-impregnated, and randomly selected specimens from a semi-pure culture. Measurements in μm. CV, coefficient of variation in %; M, median; Max, maximum; Mean, arithmetic mean; Min, minimum; *n*, number of specimens investigated; SD, standard deviation; SE, standard error of mean.

[b]As explained in ''Materials and methods''.

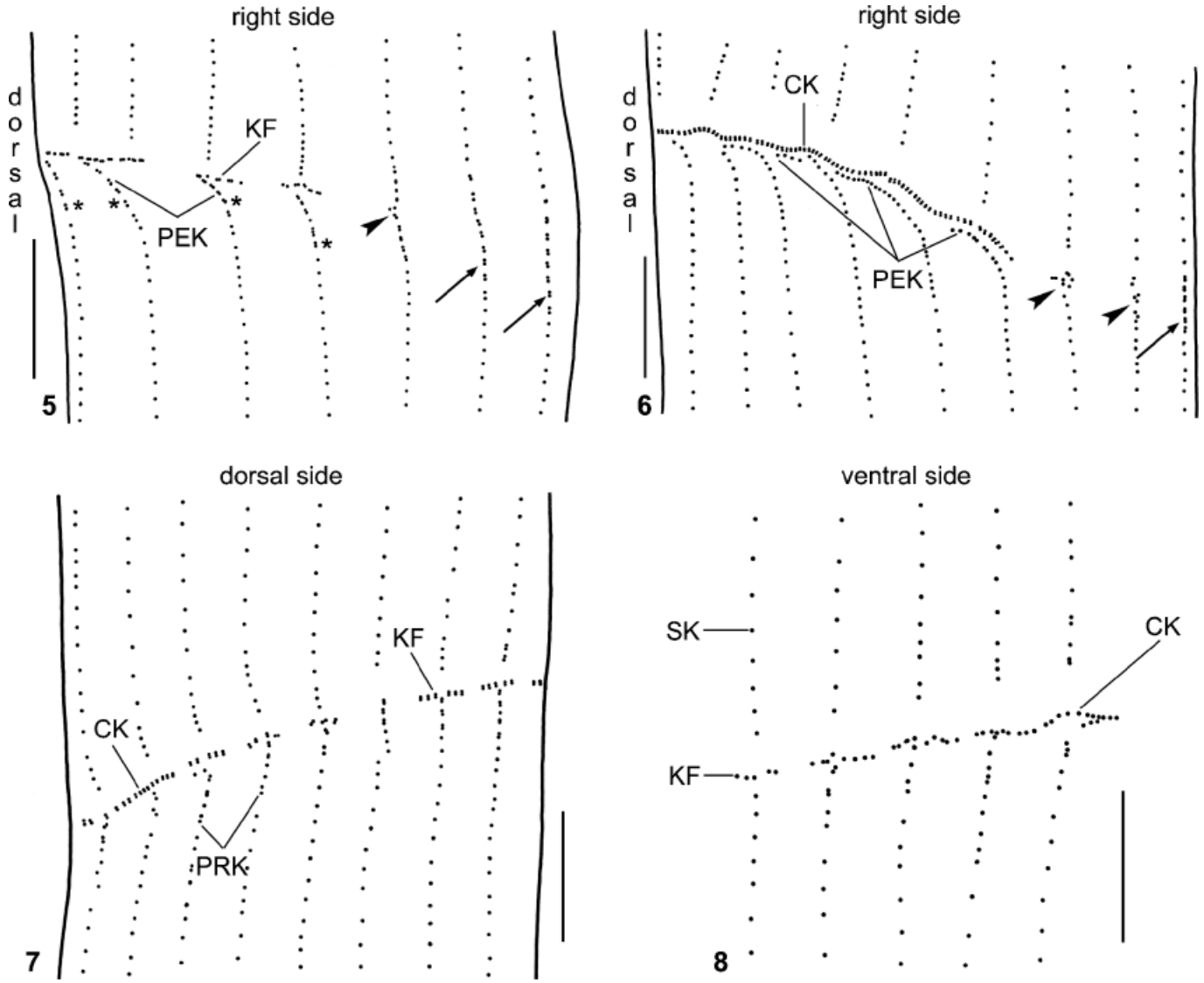


Fig. **5–8.** *Dileptus terrenus*, ciliary pattern of early dividers after protargol impregnation. **5, 6.** Right side views, showing intrakinetal proliferation of basal bodies in the lateral kineties (arrows), following a dorso-ventral gradient. Then, the new dikinetids form minute anarchic fields (arrowheads) that develop to kinetofragments producing the circumoral kinety. A second round of basal body production in the anterior region of the opisthe's right side kineties generates the monokinetidal perioral kinetofragments. During this process, dikinetid-like kinetids are recognizable (asterisks), which, however, are just divided basal bodies. The productive portion, where basal bodies become very narrowly spaced, curves dorsally (Fig. 6) and separates from the ciliary rows to form the perioral kinety (see Fig. 9, 11). **7, 8.** The oral kinetofragments are dikinetidal on the dorsal side, while monokinetidal on the ventral side; the kinetofragments are arranged transversely to the opisthe's ciliary rows, forming a T-shaped pattern. CK, circumoral kinety; KF, circumoral kinetofragments; PEK, perioral kinetofragments; PRK, preoral kinetofragments; SK, somatic kinety. Scale bars 10 μm.

dividers, the bud becomes more distinct due to the body's constriction, which occurs in a dorsoventral gradient (Fig. 20, 21). Slightly before separation, the daughter cells are connected with the broadly rounded posterior end of the proter and the developing oral bulge opening of the opisthe (Fig. 13–15, 21). Very early post-divisional opisthes have a characteristic triangular shape because the parental acute posterior body third is maintained and the proboscis extends along the anterior body end hardly projecting dorsally, resembling the oral bulge of ''polar'' haptorids (Fig. 31). Late opisthe post-dividers have a considerably shorter proboscis (50 vs. 94 μm) and a shorter (170 vs. 280 μm) and stouter (4.0:1 vs. 5.9:1) body than morphostatic cells. Thus, post-divisional development is associated with intense growth and stretching of the proboscis, first providing the body with a spatulate (Fig. 22) and then with a dileptid appearance (Fig. 32–34). In contrast, post-divisional proters are rather similar to morphostatic specimens because no changes occur in the parental oral apparatus, the somatic ciliature, and the dorsal brush. However, they are easily distinguished from morphostatic cells by the broadly rounded posterior end (vs. acute posterior third; Fig. 27–30). Further, they differ from morphostatic cells by the shorter (~208 vs. 280 μm) and stouter body (4.4:1 vs. 5.9:1) as well as by the proportion of body and proboscis length (46% vs. 34%), while the length of the proboscis (95 vs. 94 μm) is quite similar (Table 1). Thus, the proter post-divisional development is associated mainly with intense growth and patterning of the trunk.

*Stomatogenesis.* Stomatogenesis of *Dileptus* includes three main processes: the production of the circumoral kinety, the peri-

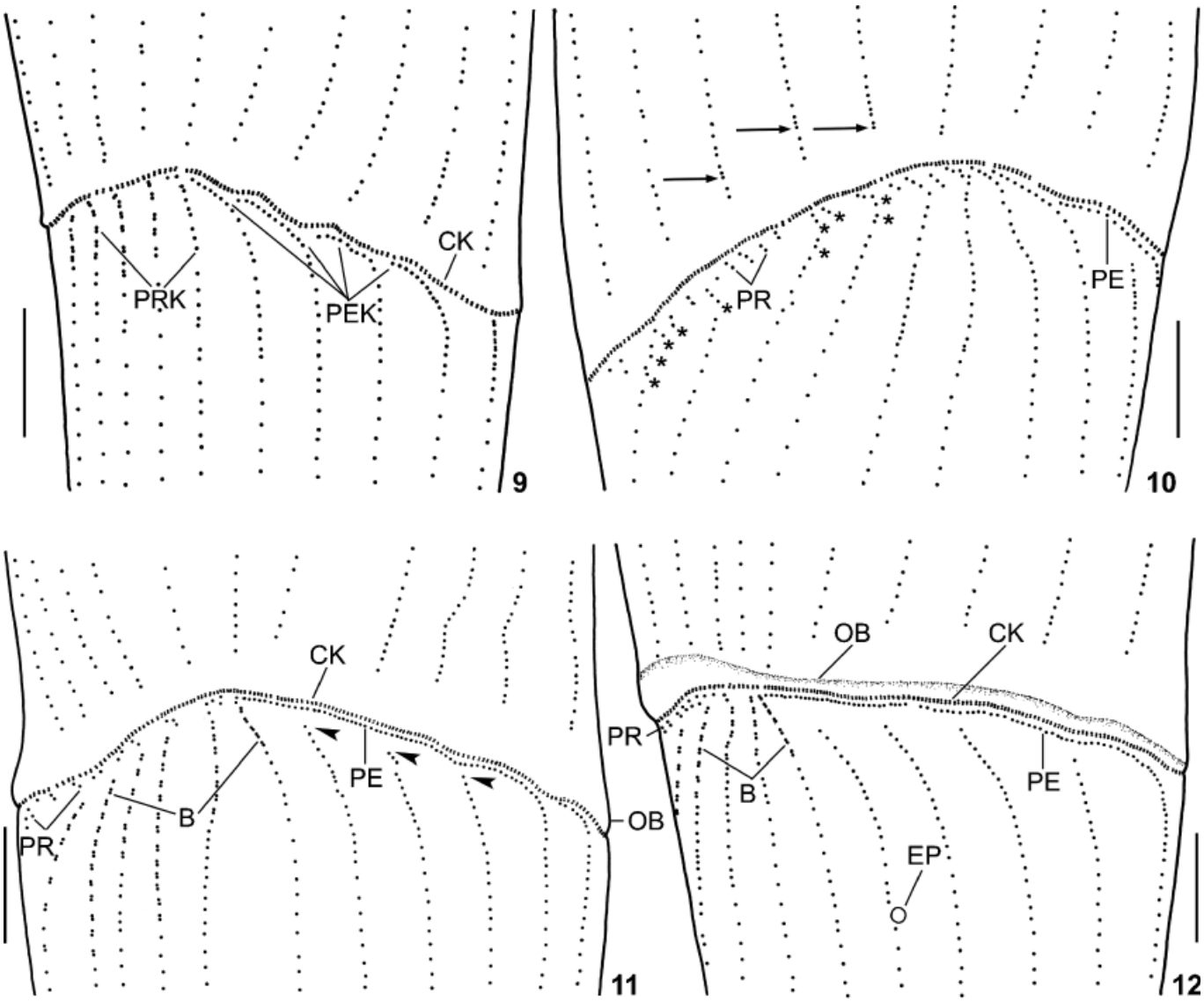


Fig. **9–12.** *Dileptus terrenus*, ciliary pattern of mid-dividers after protargol impregnation. **9.** Dorsal view of an early mid-divider, showing production of preoral kinetofragments in five dorsal ciliary rows. Like the perioral kinetofragments, they are generated by a second round of basal body production. During this process, dikinetid-like kinetids become recognizable caused, however, by just divided basal bodies (monokinetids). **10.** Mid-divider patterning preoral kineties: the individual preoral kinetofragments split into about four preoral kineties that migrate rightwards along the circumoral kinety (asterisks). Arrows mark intrakinetal proliferation of somatic kinetids, producing typical triads. **11.** A third round of basal body proliferation occurs on the dorsal side after the production of the preoral kineties and produces the multi-rowed dorsal brush. Arrowheads mark sites where the curved anterior portions of the opisthe's right side ciliary rows detached and fused to the new perioral kinety (cp. Fig. 9). **12.** Late mid-divider with developing oral bulge. The preoral kineties are arranged almost perpendicularly to the new circumoral kinety. B, dorsal brush; CK, circumoral kinety; EP, excretory pore of a contractile vacuole; OB, oral bulge; PE, perioral kinety; PEK, perioral kinetofragments; PR, preoral kineties; PRK, preoral kinetofragments. Scale bars 10 μm.

oral kinety, and the preoral kineties. The anarchic fields, which will become circumoral kinetofragments, are generated in all somatic kineties during the first round of basal body proliferation, while the perioral and preoral kinetofragments are formed during the second round, which occurs only in the right lateral and dorsal kineties, respectively (Fig. 54).

*Development of circumoral kinety and oral basket.* Division commences with the production of basal bodies in the dorsal kineties slightly posterior to the mid-body, causing the proter to be longer than the opisthe by a ratio of 1.3:1 (Table 1). Later on, proliferation of basal bodies commences in the ventral kineties slightly posterior to the level in the dorsal region, resulting in a slightly oblique division furrow (Fig. 5, 6). The newly produced basal bodies form minute anarchic fields following a dorso-ventral gradient (Fig. 5, 6, arrowheads); then they arrange transversely to the main body axis (Fig. 5, 7), forming short circumoral kinetofragments that grow and unite with the fragments from the other kineties to generate the circumoral kinety (Fig. 5–7). Interestingly, the circumoral kinetofragments originating from the right and left as well as the dorsal kineties are composed of dikinetids (Fig. 5–7), while those produced by the ventral kineties consist of monokinetids (Fig. 8). This peculiarity leads to the composite character of the circumoral kinety: oral dikinetids in the proboscis, but oralized somatic monokinetids around the oral bulge opening.

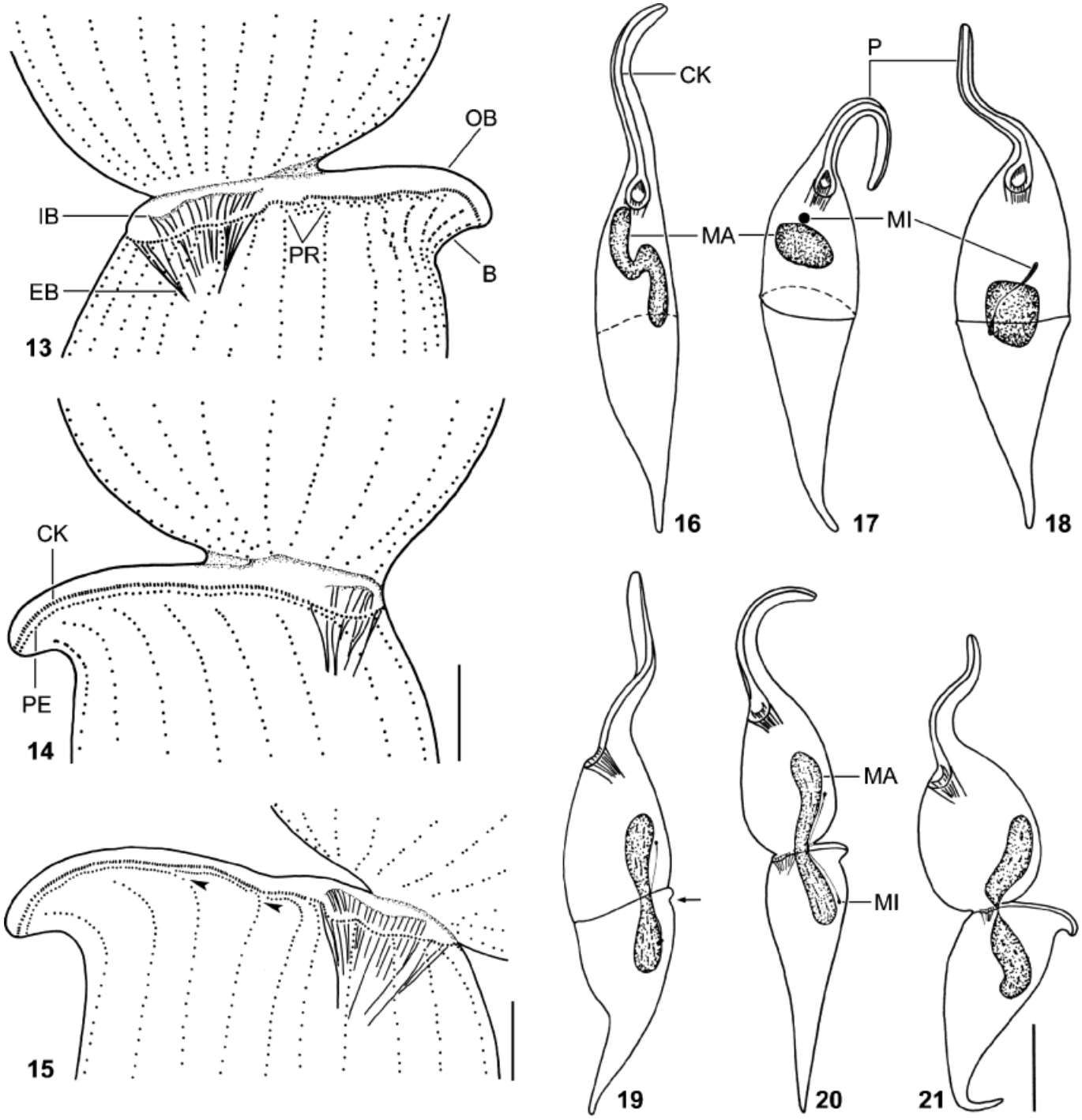


Fig. **13–21.** *Dileptus terrenus*, ciliary pattern of very late dividers (13–15) and body as well as nuclear changes in an early divider (16), mid-dividers (17, 18), and late dividers (19–21) after protargol impregnation. **13, 14.** Left and right side view of a late divider, showing the only slightly projecting proboscis and the developing oral basket as well as oral bulge opening. **15.** Right side view of a very late divider. The proboscis becomes more distinct due to the body's constriction, which occurs in a dorsoventral gradient. Arrowheads mark sites where the densely ciliated curved anterior portion of the opisthe's right side ciliary rows will detach to contribute to the perioral kinety. **16.** Ventral view of an early divider, showing the elongating macronucleus, which becomes S-shaped. **17, 18.** Ventral views of mid-dividers with condensed macronucleus and dividing micronucleus. **19–21.** Lateral views of late and very late dividers, showing division of macronucleus and micronucleus. Arrow denotes proboscis bud. B, dorsal brush; CK, circumoral kinety; IB, internal basket; EB, external basket; MA, macronucleus; MI, micronucleus; OB, oral bulge; P, proboscis; PE, perioral kinety; PR, preoral kineties. Scale bars 10 μm (Fig. 13–15) and 50 μm (Fig. 16–21).

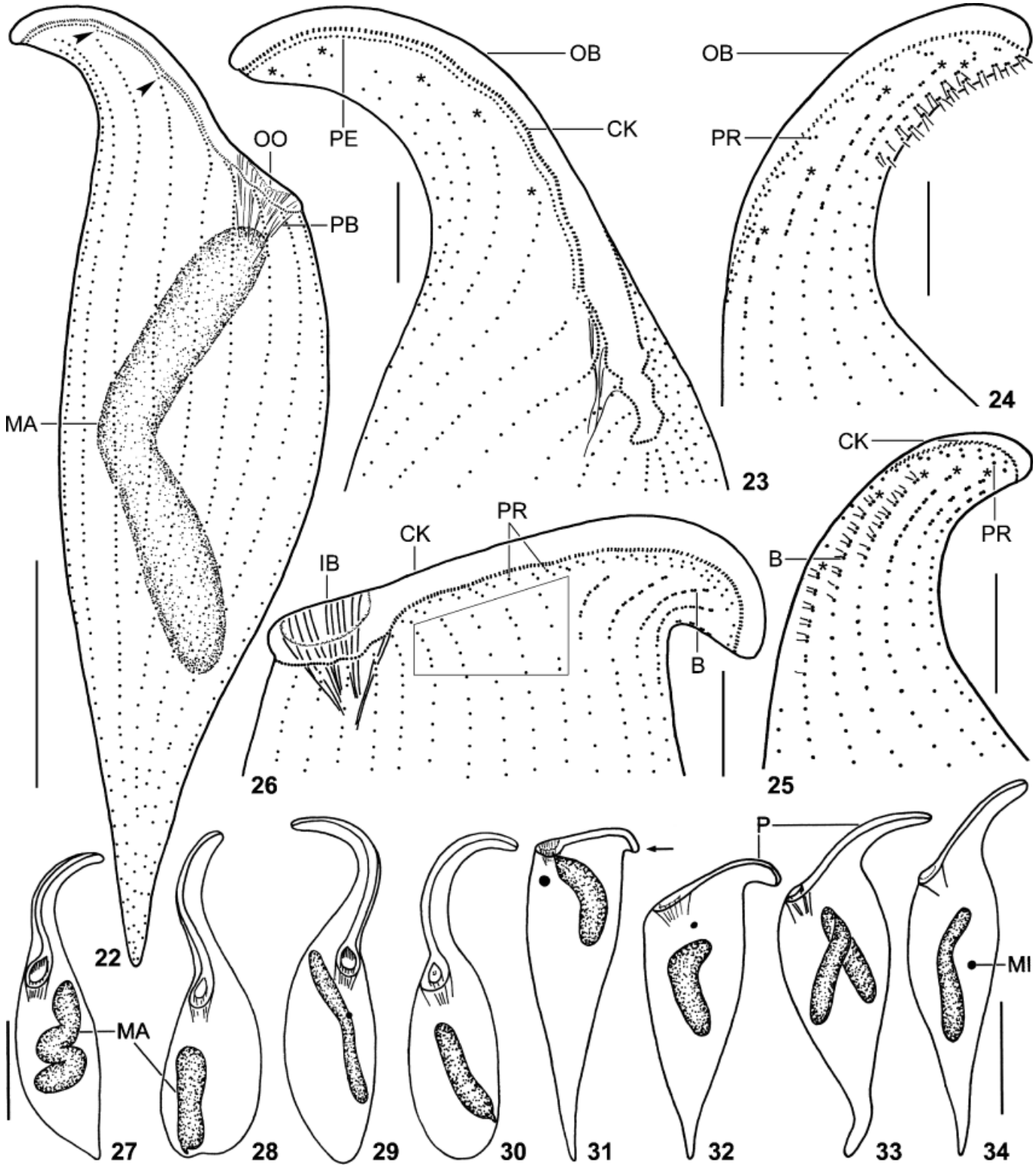


Fig. **22–34.** *Dileptus terrenus*, ciliary pattern of opisthe post-dividers (22–26) as well as body and nuclear changes in proter (27–30) and opisthe (31–34) post-dividers after protargol impregnation. **22.** Right side view of ciliary pattern, oral basket, and nuclear apparatus of an early opisthe post-divider. Arrowheads denote small irregularities in the new perioral kinety. **23.** Ventrolateral view of a malformed specimen with spathidiid circumoral kinety. Asterisks mark gradually shortened right side somatic kineties. **24, 25.** Dorsolateral views of proboscis. The dorsal brush consists of six, staggered rows (asterisks). **26.** Left side view of anterior body portion of a very early post-divider. The preoral kineties, each composed of two to three basal bodies, form minute rows arranged perpendicularly to obliquely to the circumoral kinety. The region, where new preoral kineties are possibly produced post-divisionally, is surrounded by an irregular quadrilateral. **27–30.** Variability of body shape and size as well as of the nuclear apparatus in proter post-dividers. Drawn to scale. **31–34.** Development of body shape and nuclear apparatus in opisthe post-dividers. Arrow in Fig. 31 denotes the minute, indistinctly projecting proboscis of a very early post-divider. Drawn to scale. B, dorsal brush; CK, circumoral kinety; IB, internal basket; MA, macronucleus; MI, micronucleus; OB, oral bulge; OO, oral bulge opening; P, proboscis; PB, pharyngeal basket; PE, perioral kinety; PR, preoral kineties. Scale bars 10 μm (Fig. 23–26), 30 μm (Fig. 22), and 50 μm (Fig. 27–34).

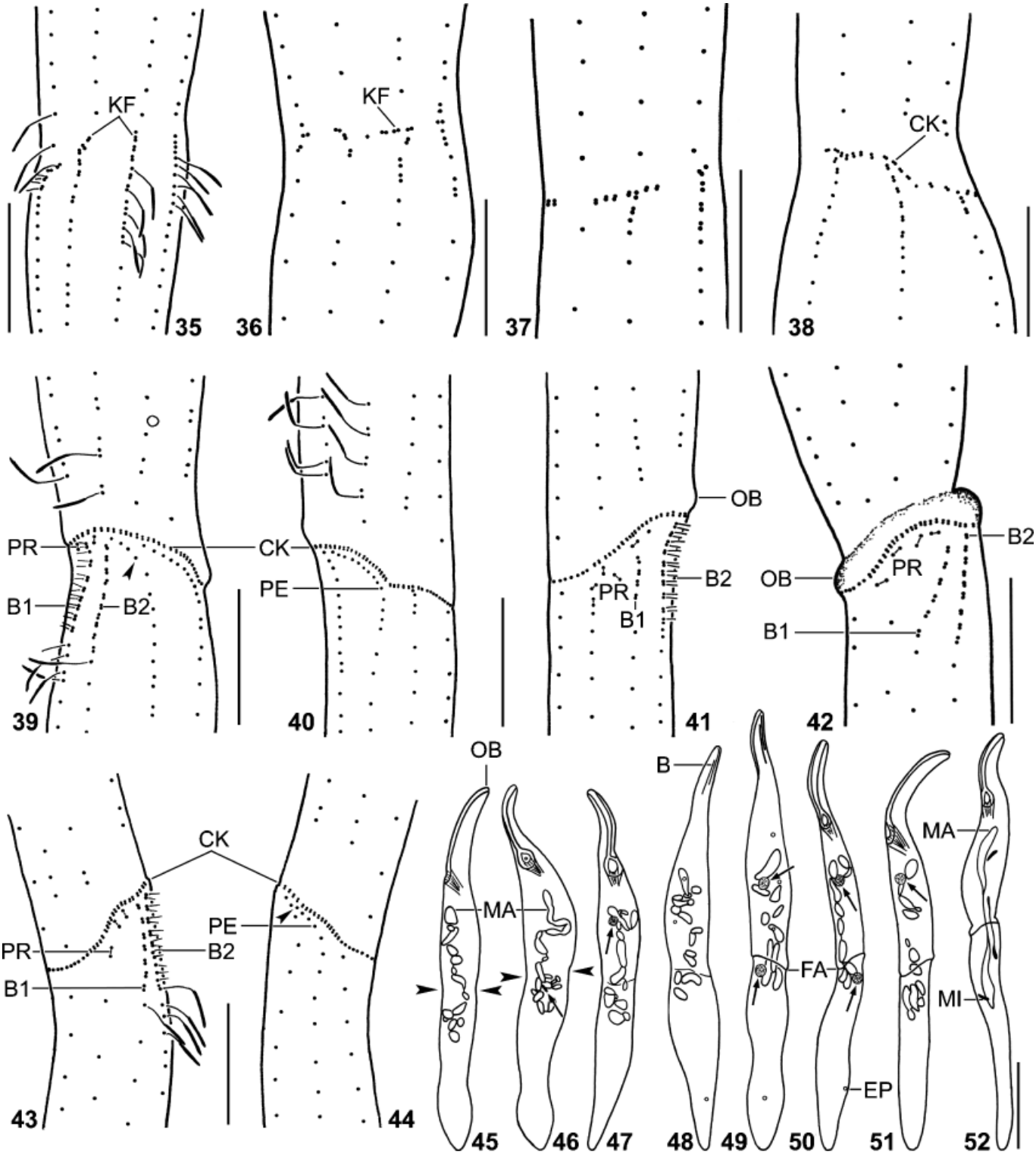


Fig. **35–52.** *Pseudomonilicaryon brachyproboscis*, ciliary pattern (35–44) and body as well as nuclear changes (45–52) in various dividers after protargol impregnation. **35–38.** Right side (35) and dorsal (36–38) views of very early and early dividers developing circumoral kinetofragments which spread both to right and left, forming a T-shaped pattern. **39, 40, 44.** Dorsolateral and right side views of mid-dividers. The perioral kinety is produced from kinetofragments developing in the anterior portion of two right side ciliary rows, but the more dorsally located kinety contributes with fewer kinetids (arrowheads) than the more ventrally located one. **41–43.** Left side views of mid-dividers developing preoral kineties (connected by lines) by fragmenting the anterior portion of the first kinety right of the dorsal brush. Possibly, brush row 1 is sometimes also involved in this process (Fig. 41). **45–52.** Body and nuclear changes in very early and early dividers as well as mid-dividers. Very early dividers are indented in the prospective fission area (arrowheads). Arrows denote the micronuclei which are narrowly ellipsoidal in very early dividers but soon become globular. Drawn to scale. CK, circumoral kinety; B(1, 2), dorsal brush (row 1, 2); EP, excretory pore of contractile vacuole; FA, fission area; KF, circumoral kinetofragments; MA, macronucleus; MI, micronucleus; OB, oral bulge; PE, perioral kinety; PR, preoral kineties. Scale bars 10 μm (Fig. 35–44) and 30 μm (Fig. 45–52).

The new oral bulge opening and the oral basket become distinct in late dividers. The nematodesmata of the external basket originate exclusively from the oralized somatic monokinetids surrounding the oral bulge opening. The rods of the internal basket, which are embedded in the developing oral bulge, are formed by the laminar transverse microtubule arrays originating also from the oralized somatic monokinetids (Grain and Golińska 1969; Fig. 13–15).

*Development of perioral kinety.* On the right side, the anterior portion of about seven opisthe's ciliary rows elongates by a second round of basal body proliferation (Fig. 5, asterisks) and curves dorsally along the growing circumoral kinetofragments (Fig. 6, 9). Later on, the curved portions detach from the ciliary rows—except for the ventralmost ciliary row, which is thus continuous with the perioral kinety in morphostatic specimens—and fuse to a continuous perioral kinety with narrowly spaced basal bodies (Fig. 11, 12, 14). This process begins in mid-dividers, identified as specimens with condensed macronucleus, and is completed in very late dividers or post-divisionally, as evident from small irregularities and/or ciliary rows still connected to the new perioral kinety (Fig. 22). In post-dividers, the perioral kinety elongates concomitantly with proboscis growth by intrakinetal proliferation of basal bodies.

*Development of preoral kineties.* During the second round of basal body proliferation, rather long preoral kinetofragments are produced in the anterior portion of about six dorsal kineties (Fig. 9). In mid-dividers, the individual kinetofragments split into about four minute portions, each consisting of two to three kinetids, which migrate rightwards to form the preoral kineties (Fig. 10, asterisks). Taking the averages of split short rows (4; Fig. 10) and the ciliary rows producing fragments (6; Fig. 9), there are about 24 preoral kineties, which is half the number found in morphostatic cells (Table 1). Thus, the other half of the preoral kineties must be generated post-divisionally during growth of the proboscis, possibly by proliferation from the existing preoral kineties or by migrating kinetids from the anterior end of those somatic kineties that terminate left of the oral bulge opening (Fig. 26).

*Development of somatic ciliature and dorsal brush.* The mature ciliary pattern of *Dileptus* develops post-divisionally and includes three specific processes in middle-sized and large species: (1) the formation of a suture along the right branch of the circumoral kinety; (2) the formation of a barren stripe along the left branch of the circumoral kinety; and (3) the formation of a staggered dorsal brush. The two first peculiarities may be inconspicuous or even absent in small species with $<10$ ciliary rows, such as *P. brachyproboscis*.

The formation of the right side suture occurs in that only the more dorsally located kineties extend to the top of the proboscis, while others are gradually shortened (Fig. 22, 23, asterisks). The barren stripe along the left branch of the circumoral kinety originates in a similar way, some kineties left of the oral bulge opening do not elongate.

The formation of the dorsal brush and the staggered arrangement of the brush rows is a complex process. In late mid-dividers, a third round of basal body proliferation produces the dikinetidal dorsal brush. This is an intense process which occurs in the six or seven dorsal ciliary rows that have produced the preoral kineties (Fig. 11, 12). Initially, all brush rows abut on the newly formed preoral kineties and thus do not have a staggered pattern (Fig. 12). In this stage, the opisthe's dorsal brush occupies a rather large, convex area because the interkinetal distance between the brush rows is only slightly smaller than that between ordinary ciliary rows (Fig. 11, 12). In late dividers and post-dividers, the elongation of the proboscis causes a decrease in the interkinetal distances (Fig. 13) and the staggered pattern of the brush rows in that they gradually elongate from ventral to dorsal (Fig. 24–26). A distinct

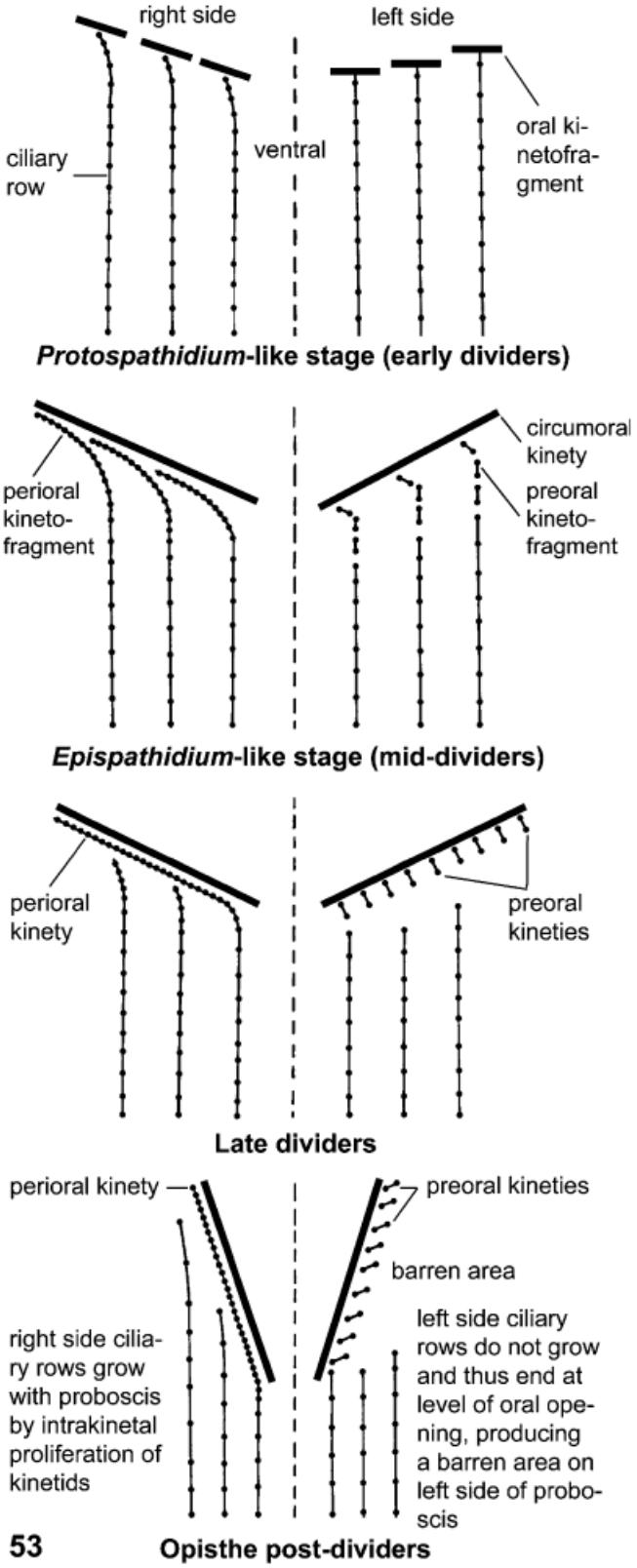


Fig. **53.** Supposed evolution of the dileptid ciliary pattern from a protospathidiid ancestor.

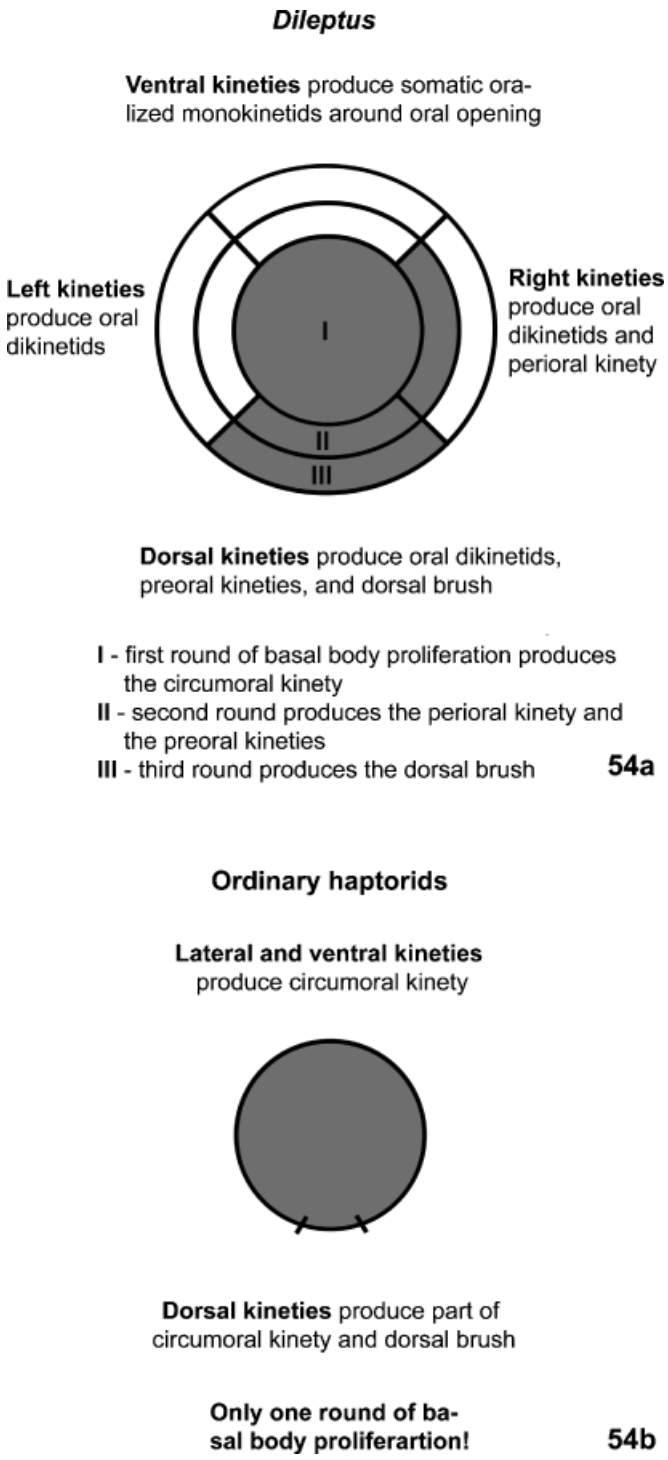


Fig. **54.** Defining ventral, lateral, and dorsal ciliary rows according to their ontogenetic activities in dileptids (a) and haptorids (b) in general. The productive regions are shaded gray.

increase in the number of brush dikinetids occurs only in late post-dividers, where basal bodies with short bristles and non-ciliated dikinetids appear in the posterior region of the brush (Fig. 24, 25). Now, the entire dorsal surface of the proboscis is covered with brush kinetids and the number of dikinetids increases from an average of 34 (n = 4) in early post-dividers to 56 in morphostatic cells (Table 1).

*Nuclear division.* In very early dividers, the macronucleus is still highly similar to that of morphostatic cells: it is cylindroidal, more or less curved, and 84 μm long on average (Table 1). Later on, the macronucleus elongates to an average length of 100 μm and becomes S- or U-shaped (Fig. 16). In mid-dividers, the macronucleus condenses to a globular, homogenously impregnated mass about 50 μm across (Fig. 17, 18). When the proboscis bud appears, the macronucleus begins to divide, becoming dumbbell shaped (Fig. 19). Then, the dividing macronucleus extends to a long rod constricting in the fission area (Fig. 20). When cell fission is finishing, the macronucleus is divided into two oblong pieces connected with their pointed ends (Fig. 21). After cell fission, the macronucleus commences to elongate and to migrate to mid-body (Fig. 27–34).

During the first stages of ontogenesis, the micronucleus increases in size from an average of 3.6 to 6.3 μm (Table 1). When the macronucleus is condensed to a globular mass, the micronucleus begins to divide becoming dumbbell shaped, with the narrowly cuneate halves connected by a fiber bundle (Fig. 18). Later on, the bundle conspicuously elongates and the daughter micronuclei become globular and homogenously impregnated (Fig. 19). In very late dividers, the micronuclei achieve the species-specific size (4 μm across), but are still connected by a long fiber (Fig. 20). During post-divisional cell growth, the micronucleus moves to mid-macronucleus (Fig. 29, 31, 32, 34).

**Observations on ontogenesis of *Pseudomonilicaryon brachyproboscis* (Fig. 35–52).** *Pseudomonilicaryon brachyproboscis* is a slender soil dileptid with a short proboscis occupying only one-fifth of body length; a moniliform macronucleus; two narrowly ellipsoidal micronuclei; and seven ciliary rows, two of which become dimorphic, staggered dorsal brush rows anteriorly (Vd'ačný and Foissner 2008b). The ontogenesis basically agrees with that of *D. terrenus* but the data are less detailed because only few dividers were found in the protargol slides. Further, the preparations are more difficult to interpret due to the low number of ciliary rows. The following peculiarities are probably genus- or species-specific: (1) early dividers display a transient indentation in the prospective fission area, just as known from several spathidiids (Fig. 45, 46, arrowheads); (2) the anterior portion of two right side ciliary rows generates the new perioral kinety, but the more dorsally located kinety contributes fewer kinetids than the more ventrally located one (1–4 vs. ~10 kinetids; Fig. 39, 40, 44, arrowheads); (3) the preoral kineties are produced by only one kinety—the first row right of the dorsal brush (Fig. 41–43); (4) the staggered pattern of the dorsal brush rows and the dimorphic arrangement of the dikinetids develop post-divisionally (Fig. 39, 41–43); and (4) the micronucleus massively changes its shape from narrowly ellipsoidal (3 × 1 μm in size) to globular (~4 μm in diam.) during early ontogenesis (cf. Fig. 46 with Fig. 47, 49–51, arrows).

## DISCUSSION

**Dileptid division mode.** Only Golińska (1972, 1995) provided detailed data on ontogenesis of dileptids, focusing on the ciliary pattern of dividers. The present study is the only one that investigated concomitantly development of cell shape, nuclear apparatus, and ciliary pattern during and, especially, after binary fission where the typical *Dileptus* shape is generated. Ontogenesis of *D. terrenus* basically agrees with data from congeners like *D. anatinus*, *D. jonesi*, *D. margaritifer*, and *D. visscheri* (Bohatier and Kink 1977; Golińska 1972, 1995; Jones 1951), all displaying the following events: (1) cell division occurs in active (non-encysted) condition; (2) the macronucleus is homomeric; (3) stomatogenesis is holotelokinetal and the parental oral apparatus does not reorganize; (4) small anarchic fields, formed at the an-

terior end of the broken ciliary rows, develop into circumoral kinetofragments growing and uniting as the circumoral kinety; (5) the perioral kinety is formed by the alignment of the densely ciliated anterior region of the right side ciliary rows; (6) the preoral kineties are produced by splitting of the anterior region of the dorsal ciliary rows into several minute portions that migrate rightwards along the circumoral kinety; (7) the dorsal brush develops very late, after the production of the preoral kineties in late mid-dividers; and (8) the proboscis basically maturates post-divisionally.

Some variation occurs in the dileptid division, just as in spathidiids (Foissner and Xu 2007). Main deviations comprise: (1) the presence/absence of a transient indentation in the prospective fission area—present in *P. brachyproboscis* but absent in *D. terrenus*; (2) the presence/absence of macronucleus condensation in mid-dividers—the nodules fuse to a mass in *P. brachyproboscis*, while they divide individually in *D. jonesi* (Jones 1951); and (3) size and shape changes of the micronucleus during early division—the micronucleus shows a 2-fold size increase but no shape changes in *D. terrenus*, while it becomes globular due to width increase in *P. brachyproboscis*. Certainly, many more peculiarities will be found when more species have been investigated.

**Comparative ontogenesis.** Ontogenesis of dileptids is much more complex than in other haptorids, displaying many peculiarities and reaching a complexity comparable to that found in ''higher'' ciliates. In dileptids, the circumoral kinetofragments are formed via small anlagen fields (Golińska 1995; present study), a widespread mode among ciliates but as yet not found in other haptorids (Foissner 1996). Further, the circumoral kinetofragments of dileptids are transversely arranged from the beginning of their formation, while they are longitudinally oriented when formed and then rotate clockwise to become horizontally arranged in spathidiids (Foissner and Xu 2007). Interestingly, a similar process occurs during the formation of the perioral kinety. A further peculiarity of *Dileptus* ontogenesis is the complex structure of the circumoral kinety: the ventral kinetofragments are composed of oralized somatic monokinetids, while the kinetofragments originating from the lateral and dorsal kineties are composed of oral dikinetids (Golińska 1995; present study). On the other hand, the circumoral kinetofragments of ordinary haptorids are exclusively dikinetidal (Foissner 1996), except for the Enchelyina, which lack oral dikinetids at all (Foissner and Foissner 1988). Unlike all haptorids investigated so far (Berger, Foissner, and Adam 1983; Foissner 1996; Foissner and Xu 2007), dileptids develop the dorsal brush as the last ciliary structure, that is, in late mid-dividers.

Another peculiarity in the division process of *Dileptus* is the three rounds of basal body production that generate the opisthe's oral and somatic ciliary pattern (Fig. 54): the first round, where all kineties are involved as in ordinary haptorids, generates the circumoral kinety; the second round, where only part of the kineties is involved, generates the perioral and preoral ciliature; and the third round, which generates the dorsal brush, is limited to a few dorsal kineties. We define ''round'' in contrast to ''continuous,'' where the ciliary structures are produced in a steady process from a bulk of basal bodies usually generated in the early division stages, for instance, from an ''anarchic field'' or intrakinetally during the whole divisional and post-divisional process. Typical examples are tetrahymenids (Foissner 2003), while hypotrichs are typical ''rounders,'' generating their adoral membranelles by three rounds of basal body production (Jerka-Dziadosz 1981).

As yet, we do not know the phylogenetic significance of the number of rounds basal bodies are generated. Possibly, the ''one round'' pattern is the plesiomorphic state because it is most widespread occurring, for instance, in karyorelictids (Foissner and Al-Rasheid 1999), metopids (Foissner and Agatha 1999), and tetrahymenids (Foissner 2003). At the present state of knowledge, the number of rounds can be used pragmatically as a measure of division complexity.

At first glance, the complex ontogenesis of dileptids appears to be caused by the proboscis. However, two observations—the unique formation of the circumoral kinetofragments and the late genesis of the dorsal brush—suggest that this is only part of the truth. Both peculiarities are obviously independent of spatial constraints and the presence/absence of a proboscis.

**Phylogeny.** Several data suggest a close relationship of spathidiids and dileptids (Vd'ačný and Foissner 2008a; present study), but it is not known whether the dileptids evolved from a *Spathidium*-like ancestor by the formation of a proboscis or the spathidiids lost the proboscis of a *Dileptus*-like ancestor (Xu and Foissner 2005). There are five observations that suggest a spathidiid ancestor of dileptids (Fig. 53): (1) the formation of various spathidiid body shapes and ciliary patterns during ontogenesis and conjugation of *Dileptus* (Vd'ačný and Foissner 2008a; present study); (2) the composite character of the dileptid circumoral kinety—oral dikinetids in the proboscis, while oralized somatic monokinetids around the oral bulge opening, which is either a plesiomorphic state inherited from an enchelyine ancestor or a highly derived state; (3) the location of the oral bulge opening at the base of the proboscis is likely an apomorphic state, as indicated by the detailed electron microscopical investigations of Golińska (1995); (4) the post-divisional maturation of the proboscis, which indicates its apomorphic state; and (5) the short dorsal process, resembling a proboscis, found in several spathidiids (Foissner and Xu 2007). On the other hand, there are two features favoring a dileptid ancestor of the spathidiids: (1) molecular analyses place *Dileptus* basal to the other haptorids but the data are still too incomplete for a firm conclusion (Strüder-Kypke et al. 2006); and (2) the spathidiid adesmokinety-like fragments could be homologous to the preoral kineties of *Dileptus* (Xu and Foissner 2005). Independent of the ancestor, the various peculiarities of the dileptid morphology and ontogenesis could indicate a long, independent evolution, probably justifying the ordinal rank suggested by Jankowski (1980).

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## LITERATURE CITED

Corliss, J. O. 1979. The Ciliated Protozoa. Characterization, Classification and Guide to the Literature. 2nd ed. Pergamon Press, Oxford.

Berger, H., Foissner, W. & Adam, H. 1983. Morphology and morphogenesis of *Fuscheria terricola* n. sp. and *Spathidium muscorum* (Ciliophora: Kinetofragminophora). *J. Protozool.*, **30**:529–535.

Bohatier, J. & Kink, J. 1977. Etude des synthèses protéiques au cours des processus morphogénétiques de division et de régénération chez *Dileptus anser*: action de la cycloheximide. *Protistologica*, **13**:509–528.

Dragesco, J. 1963. Révision du genre *Dileptus*, Dujardin 1871 (Ciliata Holotricha) (systématique, cytologie, biologie). *Bull. biol. Fr. Belg.*, **97**:103–145.

Foissner, W. 1981. Morphologie und Taxonomie einiger neuer und wenig bekannter kinetofragminophorer Ciliaten (Protozoa: Ciliophora) aus alpinen Böden. *Zool. Jb. Syst.*, **108**:264–297.

Foissner, W. 1984. Infraciliatur, Silberliniensystem und Biometrie einiger neuer und wenig bekannter terrestrischer, limnischer und mariner Ciliaten (Protozoa: Ciliophora) aus den Klassen Kinetofragminophora, Colpodea und Polyhymenophora. *Stapfia*, **12**:1–165.

Foissner, W. 1991. Basic light and scanning electron microscopic methods for taxonomic studies of ciliated protozoa. *Europ. J. Protistol.*, **27**: 313–330.

Foissner, W. 1996. Ontogenesis in ciliated protozoa, with emphasis on stomatogenesis. *In*: Hausmann, K. & Bradbury, B. C. (ed.), Ciliates: Cells as Organisms. Fischer Verlag, Stuttgart. **46**:95–177.

Foissner, W. 1997. Faunistic and taxonomic studies on ciliates (Protozoa, Ciliophora) from clean rivers in Bavaria (Germany), with descriptions of new species and ecological notes. *Limnologica*, **27**:179–238.

Foissner, W. 2003. Morphology and ontogenesis of *Lambornella trichoglossa* nov. spec., a new tetrahymenid ciliate (Protozoa, Ciliophora) from Brasilian tank bromeliads (Bromeliaceae). *Europ. J. Protistol.*, **39**:63–82.

Foissner, W. & Agatha, S. 1999. Morphology and morphogenesis of *Metopus hasei* Sondheim, 1929 and *M. inversus* (Jankowski, 1964) nov. comb. (Ciliophora, Metopida). *J. Eukaryot. Microbiol.*, **46**:174–193.

Foissner, W. & Al-Rasheid, K. A. S. 1999. Ontogenesis in a trachelocercid ciliate (Ciliophora: Karyorelictea), *Sultanophrys arabica*, with an account of evolution at the base of the ciliate tree. *Acta Protozool.*, **38**:273–290.

Foissner, W. & Foissner, I. 1988. The fine structure of *Fuscheria terricola* Berger et al., 1983 and a proposed new classification of the subclass Haptoria Corliss, 1974 (Ciliophora, Litostomatea). *Arch. Protistenk.*, **135**:213–235.

Foissner, W. & Xu, K. 2007. Monograph of the Spathidiida (Ciliophora, Haptoria). Vol. I: Protospathidiidae, Arcuospathidiidae, Apertospathulidae. Springer, Dordrecht.

Foissner, W., Agatha, S. & Berger, H. 2002. Soil Ciliates (Protozoa, Ciliophora) from Namibia (Southwest Africa) with emphasis on two contrasting environments, the Etosha region and the Namib Desert. *Denisia*, **5**:1–1459.

Fryd-Versavel, G., Iftode, F. & Dragesco, J. 1975. Contribution a la connaissance de quelques ciliés gymnostomes II. Prostomiens, pleurostomiens: morphologie, stomatogenèse. *Protistologica*, **11**:509–530.

Golińska, K. 1972. Studies on stomatogenesis in *Dileptus* (Ciliata, Holotricha) in the course of division processes. *Acta Protozool.*, **9**:283–297.

Golińska, K. 1991. Cortical organellar complexes, their structure, formation, and bearing upon cell shape in a ciliate, *Dileptus*. *Protoplasma*, **162**:160–174.

Golińska, K. 1995. Formation and orientation of skeletal elements during development of oral territory in a ciliate, *Dileptus*. *Acta Protozool.*, **34**:101–113.

Grain, J. & Golińska, K. 1969. Structure et ultrastructure de *Dileptus cygnus* Claparède et Lachmann, 1859, Cilié Holotriche Gymnostome. *Protistologica*, **5**:269–291.

Huber-Pestalozzi, G. 1945. Neue Planktonorganismen im Zürichsee. *Paradileptus conicus* Wenrich und *Paradileptus ovalis* nova spec. (3. Mitteilung). *Vjschr. naturf. Ges. Zürich*, **90**:120–126.

Jankowski, A. W. 1980. Conspectus of a new system of the phylum Ciliophora. *Proc. Acad. Sci. USSR*, **94**:103–121. (in Russian with English title translation).

Jerka-Dziadosz, M. 1981. Ultrastructural study on development of the hypotrich ciliate *Paraurostyla weissei*. II. Formation of the adoral zone of membranelles and its bearing on problems of ciliate morphogenesis. *Protistologica*, **17**:67–81.

Jones, E. E. Jr. 1951. Encystment, excystment, and the nuclear cycle in the ciliate *Dileptus anser*. *J. Elisha Mitchell scient. Soc.*, **67**:205–217.

Kahl, A. 1931. Urtiere oder Protozoa I: Wimpertiere oder Ciliata (Infusoria) 2. Holotricha außer den im 1. Teil behandelten Prostomata. *Tierwelt Dtl.*, **21**:181–398.

Kink, J. 1976. A localized region of basal body proliferation in growing cells of *Dileptus visscheri* (Ciliata, Gymnostomata). *J. Cell. Sci.*, **20**:115–133.

Strüder-Kypke, M. C., Wright, A.-D. G., Foissner, W., Chatzinotas, A. & Lynn, D. H. 2006. Molecular phylogeny of litostome ciliates (Ciliophora, Litostomatea) with emphasis on free-living haptorian genera. *Protist*, **157**:261–278.

Vd'ačný, P. & Foissner, W. 2008a. Morphology, conjugation, and postconjugational reorganization of *Dileptus tirjakovae* n. sp. (Ciliophora, Haptoria). *J. Eukaryot. Microbiol.*, **55**:436–447.

Vd'ačný, P. & Foissner, W. 2008b. Description of four new soil dileptids (Ciliophora, Haptoria), with notes on adaptations to the soil environment. *Acta Protozool.*, **47**:211–230.

Xu, K. & Foissner, W. 2005. Morphology, ontogenesis and encystment of a soil ciliate (Ciliophora, Haptorida), *Arcuospathidium cultriforme* (Penard, 1922), with models for the formation of the oral bulge, the ciliary patterns, and the evolution of the spathidiids. *Protistology*, **4**: 5–55.

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