

Cholamonas cyrtodiopsidis gen. n., sp. n. (Cercomonadida), an Endo-commensal, Mycophagous Heterotrophic Flagellate with a Doubled Kinetid

Martin FLAVIN¹, Charles J. O'KELLY², Thomas A. NERAD³ and Gerald WILKINSON⁴

¹Laboratory of Cell Biology, National Heart, Lung and Blood Institute, National Institutes of Health, Bethesda, MD; ²Bigelow Laboratory for Ocean Sciences, McKown Point, West Boothbay Harbor, ME; ³Protistology Collection, American Type Culture Collection Manassas, VA; ⁴Zoology Department, University of Maryland, College Park, MD, U.S.A.

Summary. The name *Cholamonas cyrtodiopsidis* gen. n., sp. n., is created for a mycophagous heterotrophic flagellate isolated and cultivated from the intestine of a diopsid fly. Flagellates were subanteriorly biflagellate, uninucleate, and naked. Golgi dictyosomes were anterior to the nucleus, and a reticulate paranuclear body posterior to it. Two groups of refractile bodies were present, one at the anterior end of the cell, the other in the vicinity of the nucleus. Numerous elongate, unbranched mitochondria with tubular cristae were distributed around the periphery and parallel to the long axis of the cell. The kinetid consisted of two symmetrical subunits, each with two basal bodies (one of which was associated with a stubby flagellum), a compound microtubular root, and associated fibers and bands. Cytoskeletal microtubules emanated from the cell anterior and were not associated with any kinetid element. Feeding, on yeast, was accomplished by rapid pseudopodial action at the posterior end of the cell. *Cholamonas cyrtodiopsidis* is referred to Cercomonadida because it possesses a paranuclear body and has a kinetid architecture similar to some species of *Cercomonas*. It differs from all other cercomonads in its endo-commensal habitat, mycophagy, doubled kinetid, distribution of refractile granules and mitochondria, and minimal production of pseudopodia.

Key words: *Cercomonas*, cercomonads, *Cholamonas cyrtodiopsidis* gen. n., sp. n., kinetid architecture, paranuclear body.

Abbreviations: A - anterior basal body, AL - left anterior basal body, AR - right anterior basal body, f - food vacuole, g - Golgi, gl - apical refractile granules, gm - median refractile granules, m - mitochondria, MR - microtubular roots, MRL - left microtubular roots, MRR - right microtubular roots, n - nucleus, nu - nucleolus, p - paranuclear body (parabasal body), P - posterior basal body, PL - left posterior basal body, PR - right posterior basal body, r - ring structure, sb - striated band, w - wings, y - yeast cell.

INTRODUCTION

Cercomonads (order Cercomonadida) are common and widespread heterotrophic protists in aquatic and soil

environments (Sandon 1927, Hänel 1979, Larsen and Patterson 1990, Patterson and Zölffell 1991). Despite their frequency of occurrence, little is yet known of the biodiversity or systematic biology of these protists, and their taxonomy is uncertain.

At present, organisms referred to this group are biflagellate, tubulocristate (having mitochondria with tubular cristae; Patterson 1994) free-living, mostly

Address for correspondence: Martin Flavin, Laboratory of Cell Biology, NHLBI, National Institutes of Health, Bldg.3, Rm. B1-22, 9000 Rockville Pike, MD 20892 - 0301, U.S.A.; Fax: 301-402-1519; E-mail: flavinM@gwgate.nhlbi.nih.gov

bacterivorous flagellates or amoeboflagellates that move by gliding or swimming, take in prey by means of pseudopodia and have a conspicuous, homogeneous paranuclear body. Hairs and paraxial rods are lacking from the flagella, and cytoskeletally-defined cytostome are absent from the cell body. Patterson and Zöllffel (1991) recognized three genera: *Cercomonas* Dujardin, 1841; *Heteromita* Dujardin, 1841; *Massisteria* Larsen and Patterson, 1990. These authors suggested that *Cercobodo* Krassilstchick, 1886, is a synonym of *Cercomonas*, and that several lesser-known genera of flagellates are likewise synonyms of *Cercomonas* or *Heteromita*. Cavalier-Smith (1993) added *Discocelis* Vørs, 1988 to the order.

The morphological concepts of the order and its included genera are supported, to a greater or lesser extent, by ultrastructural studies on selected species of *Cercomonas* (Mignot and Brugerolle 1975; Mylnikov 1985, 1986a, b), *Heteromita* [(MacDonald *et al.*, 1977; Mylnikov 1985 (as *Bodomorpha* Mylnikov, 1990)], *Massisteria* (Patterson and Fenchel, 1990), and *Discocelis* (Vørs, 1988), and by molecular sequence investigations on selected species especially in the first two genera (Cavalier-Smith and Chao 1996-97).

In this report, an organism is described that has several features in common with the cercomonads that have previously been examined, but also has many novel features including an endocommensal habitat, feeding on yeast instead of bacteria, and a "doubled" kinetid. The organism is named *Cholamonas cyrtodiopsisidis* gen. n., sp. n.

MATERIALS AND METHODS

Collection, isolation and cultivation

The protist was isolated from the intestinal tract of the diopsid fly *Cyrtodiopsis dalmanni* Wiedemann, 1830, collected by GW in 1989 from Kuala Lumpur, Malaysia. This fly species is common in Indonesia, Malaysia and Thailand, where it feeds on decaying forest litter (de la Motte and Burkhardt 1993). The fly was maintained in the laboratory using pureed corn. Clonal monoprotist cultures were established by serial dilution and maintained in ATCC medium 802 (Nerad 1992) with the addition of the yeast *Saccharomyces cerevisiae* ATCC 48894; bacteria also were present. One clone was cryopreserved (Nerad and Daggett 1992, Poynton *et al.* 1995) and deposited in the American Type Culture Collection with the strain number ATCC 50325.

Light microscopy

Light microscopical observations were made on live cells and on those fixed in Bouin's fluid and stained using the quantitative protargol procedure (Lynn 1992). They were viewed on a Zeiss Axioskop light microscope equipped with bright-field, phase-contrast and differential interference contrast optics. Micrographs were made on Kodak Technical Pan 35 mm film, exposed at ISO 25 and processed with Kodak Technidol developer.

Scanning electron microscopy

Cells to be fixed were harvested from a 2-day culture. Several drops of a culture suspension were allowed to settle for 10 min on a poly-L-lysine coated Thermanox (EMS) coverslip. After removing the supernatant by touching the edge of the grid with a piece of torn filter paper, a few drops of fixative was added and left for 10 to 40 min. The fixative was either the one used for TEM (below) or Parducz's fixative; the latter yielded cells with artefactual deep longitudinal fissures. The coverslips were washed with either water or 0.1 M sodium phosphate, pH 7.4, and at once dehydrated by dipping successively for 5 min in two changes of 50, 70, and 95% ethanol and then for 5 min in three changes of 100% ethanol. Coverslips were then dipped for 10 min each in two changes of 100% hexamethyldisilazane in a vacuum hood and then placed on filter paper in a Petri dish. Coverslips were mounted on a metal stub (Pella) with silver conducting paint, and kept in a dessicator overnight. The next morning they were sputter coated for 90 seconds with gold using a Hummer-V instrument filled with argon, and then examined in a JEOL JSM 35C microscope.

Transmission electron microscopy - harvest and fixation.

Twenty ml of 8% glutaraldehyde was added to 20 ml of a 2 or 3 day (late log) culture. The mixture was centrifuged at once for 5 min at 1000 x g, the supernatant was aspirated from a small loose pellet, and the latter was resuspended by injecting, through a 22 ga. needle, 4 ml of chilled 0.1 M sodium phosphate buffer, pH 7.25, containing 3 % glutaraldehyde, 1.5% osmium tetroxide and sometimes also 2 mM MgCl₂. The suspension was allowed to rise to room temperature while standing for 45 min. The pellet was then recovered and washed twice with PBS by centrifugation for 5 min at 500 x g.

Transmission electron microscopy - enrobing and Epon embedding

After suspending the pellet in 0.9 ml of the same phosphate buffer in a conical plastic tube, 1 ml of chilled 20% bovine serum albumin was added followed by 0.3 ml of 8% glutaraldehyde, and the cold mixture was immediately centrifuge for 5 min at 5°C at 3000 rpm in a swinging bucket Sorvall RT6000B centrifuge. The tube was kept for 20 min at 37° C to solidify the contents. The bottom portion of the tube containing fixed cells was separated with a razor blade, and the contents emptied into a Petri dish containing the same phosphate buffer. The plug was minced finely with a clean razor blade. Small fragments were transferred to a scintillation bottle with 3 ml of the sodium phosphate buffer, to which 1 ml of osmium tetroxide was

added. The post-fixation was for 1h stationary at room temperature in a vacuum hood. The blocks of cells were dehydrated in a graded alcohol series followed by propylene oxide, and incubated on a rotary platform in an uncapped scintillation vial overnight in a mixture of equal volumes of propylene oxide and Epon. After again rotating overnight in Epon alone blocks were embedded in 8.5 mm gelatin capsules containing Epon and kept at 60° C for 3 days. Serial silver to pale gold sections were cut with a DDK diamond knife.

Transmission electron microscopy - staining

The serial sections were collected on formvar coated Nochnum slot grids (Pella 4518), and ten grids were placed on the Pella grid-stick which had been coated with this companies' adhesive to keep the metal edges of each grid firmly adherent. The grid stick was inserted in a small Pasteur pipette, and sections were stained for 15 min with 2% aqueous uranyl acetate. To avoid stain artifacts (possibly resulting from the two adhesives present - grid stick and Tackiwax used for sectioning), extensive washing in deionized water was necessary before the 10 min in lead citrate: besides repeated dipping, water was allowed to flow through the pipette for 15 min.

RESULTS

Trophic cells of *Cholamonas cyrtodiopsis* were cylindrical to narrowly ellipsoidal when swimming unimpeded (Fig. 1), but the cells were easily flattened under coverslip pressure (Figs. 2, 9) and could bend and twist to get around objects. No pseudopodia were observed. Cylindrical cells measured 5.0 - 8.0 μm in length and 2.0 - 5.0 μm in width at the widest point; cells deformed by flattening were shorter and wider. The cells bore two subapically-inserted, posteriorly-directed emergent flagella that usually were equal in length, 1 - 1.5 times as long as the cell body, and had tapering tips (Figs. 1, 3). The flagella contained the standard 9 + 2 axoneme except at their tips, and lacked hairs, scales and paraxial rods. The attenuated tips were seen only by SEM. Fig. 1 shows a cell with a very long tip on one flagellum, and a typical tip on the other. Nothing that could correspond to the tips has been seen by TEM; perhaps they are easily broken off.

Two flagellar stumps were present anterior to the emergent flagella (Figs. 1, 3). The stumps contained a very short axoneme consisting of nine singlet microtubules (Fig. 4).

The single nucleus was found in the anterior third of the cell, always closely associated with the flagellar insertion (Figs. 2, 3). Each interphase nucleus contained a single, prominent, central nucleolus (Fig. 5).

The Golgi apparatus, which consisted of two dictyosomes, one on either side of the kinetid, was located at

the anterior surface of the nucleus (Fig. 5). Also anterior to the nucleus, typically forming a cap at the anterior end of the cell, was a population of refractile granules (Fig. 5). These granules, not demonstrably membrane-bound and having electron-transparent contents (Figs. 5, 11), were presumably responsible for a bright apical spot in living cells which might aid identification with a light microscope.

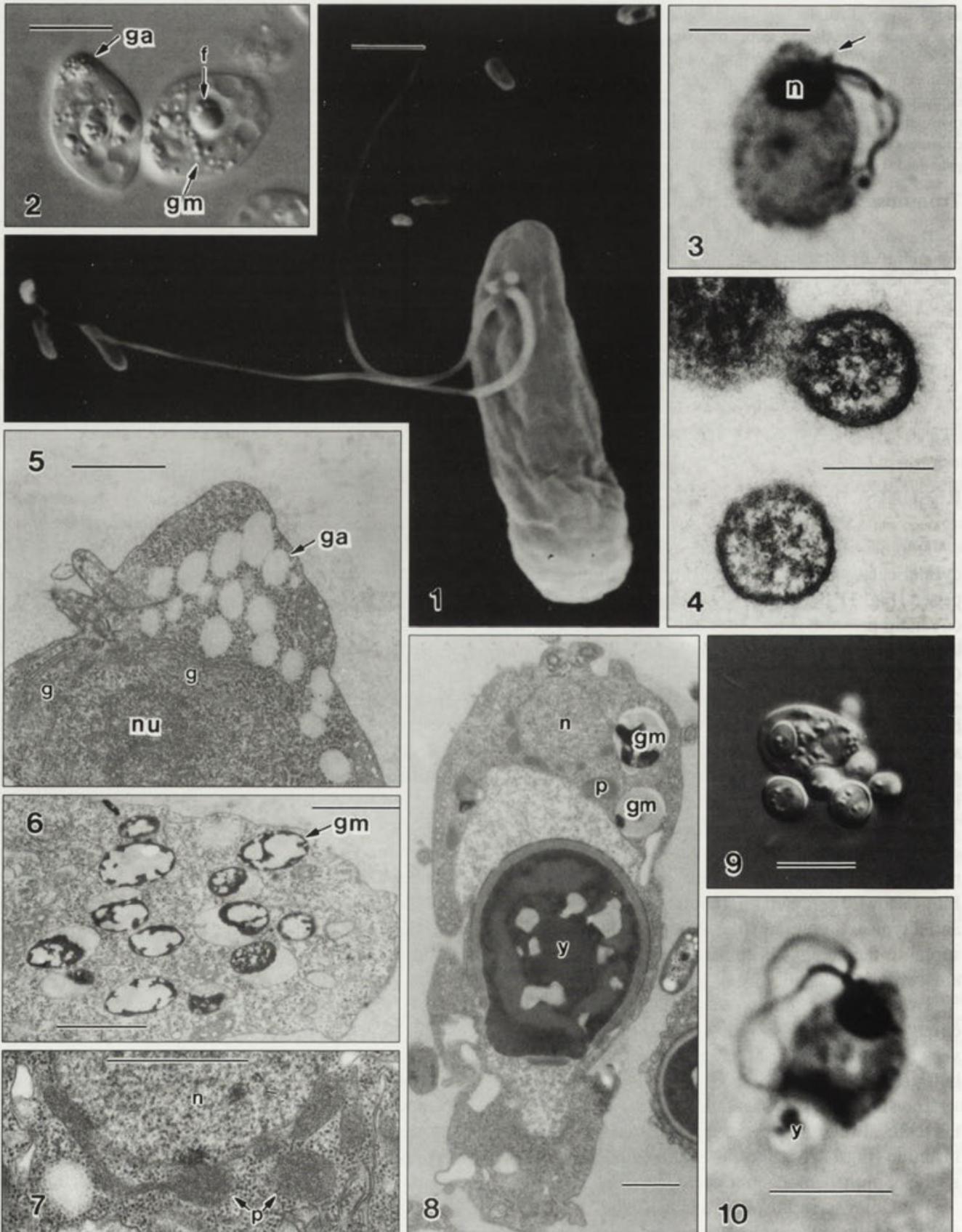
A second population of refractile granules formed a looser aggregation around and posterior to the nucleus (Figs. 2, 6). These granules were larger than the apical cap granules, were bounded by a single membrane, and had electron-dense contents (Fig. 6). Also associated with the posterior surface of the nucleus was a large and complex paranuclear body (Figs. 7, 8). Reconstruction revealed a reticulate organelle, with lobes often extending well away from the nucleus towards the posterior end of the cell.

Much of the posterior two-thirds of the cell was taken up by food vacuoles, which contained only yeast cells when full (Fig. 8). The cells did not take up, or survive on, media containing only bacteria or other species of yeast. All protist cells appeared dead within 24 h of yeast depletion, and we were not able to induce spore formation. Yeast cells taken as prey were ingested at the posterior end of the cell (Figs. 9, 10); the ingestion process took less than three seconds in the cells observed while feeding. No specialized cytostomal structures were seen.

Each cell contained around a dozen elongate, unbranched mitochondria, arranged around the periphery of the cell and extending longitudinally from the anterior to the posterior end (Figs. 11, 12). The cristae were tubular, and were preferentially arranged about the periphery of the mitochondrion, so that certain wider mitochondrial segments had crista-free central regions (Fig. 13).

No contractile vacuoles were detected.

The kinetid consisted of four basal bodies, two compound microtubular roots, and associated fibers and bands. Each of the four basal bodies was short measuring *ca* 350 μm (Fig. 14). At the distal end of each basal body, near the transition zone, wing like projections extended from the microtubule blades to the plasmalemma; at this point the C tubules of the blades were lost (Fig. 15). Transition zone structures on the two basal bodies bearing stubby flagella were not distinguishable (Fig. 14). Transition zone structures on the two basal bodies bearing emergent, functional flagella included an



indistinct basal plate (Fig. 14) and, more distally, an apparently tubular structure just inside the axonemal doublet microtubules (Figs. 14, 15).

The basal bodies were arranged in two pairs, each pair consisting of one basal body with an emergent, and one with a stubby, flagellum. The basal bodies in each pair were coplanar, separated from each other by about half a basal body diameter, and formed an angle of approximately 30 degrees (Figs. 1, 14, 16-18). The two pairs were closely juxtaposed, as if belonging to a single kinetid, but the basal bodies of the pair on the right were mounted slightly to the posterior of the elements of the pair on the left (Figs. 1, 16-18). Also, the basal bodies and flagella of the right-side pair projected towards the right side of the cell, while the basal bodies and flagella of the left-side pair projected towards the left side of the cell (Figs. 1, 16-18). No structures were observed that linked the basal bodies to each other, but the basal bodies associated with emergent flagella each had a prominent electron-dense "terminal cap" linking the proximal end of the basal body to other kinetid components, particularly the flagellar roots (Figs. 14, 19).

Two compound flagellar roots were present, one arising from underneath each of the basal body pairs and descending posteriorly along the ventral surface of the cell (Figs. 14, 16-25). The roots were identical. A striated band was associated with each root; each was inserted into electron-dense material associated with a pocket in the nuclear envelope (Figs. 14, 19-21).

At its origin, the main band of each microtubular root typically consisted of six microtubules (Fig. 21). More distally, but still in the immediate vicinity of the basal bodies, two additional microtubules were added to the outer side of the root, forming an 8-membered spline (Fig. 22). At about this point, an additional two microtubules arose from electron-dense material associated with the emergent-flagellum basal body (Figs. 22, 23) and extended posteriorly between the main spline and

the plasmalemma (Figs. 23-25). Slightly distal to the point of origin of the doublet, the outermost of the spline microtubules became separate and associated with electron-dense material (Figs. 23, 24). Microtubules subsequently disappeared from the main spline as the root descended, in a straight line, to an unknown termination point in the posterior of the cell (Fig. 25).

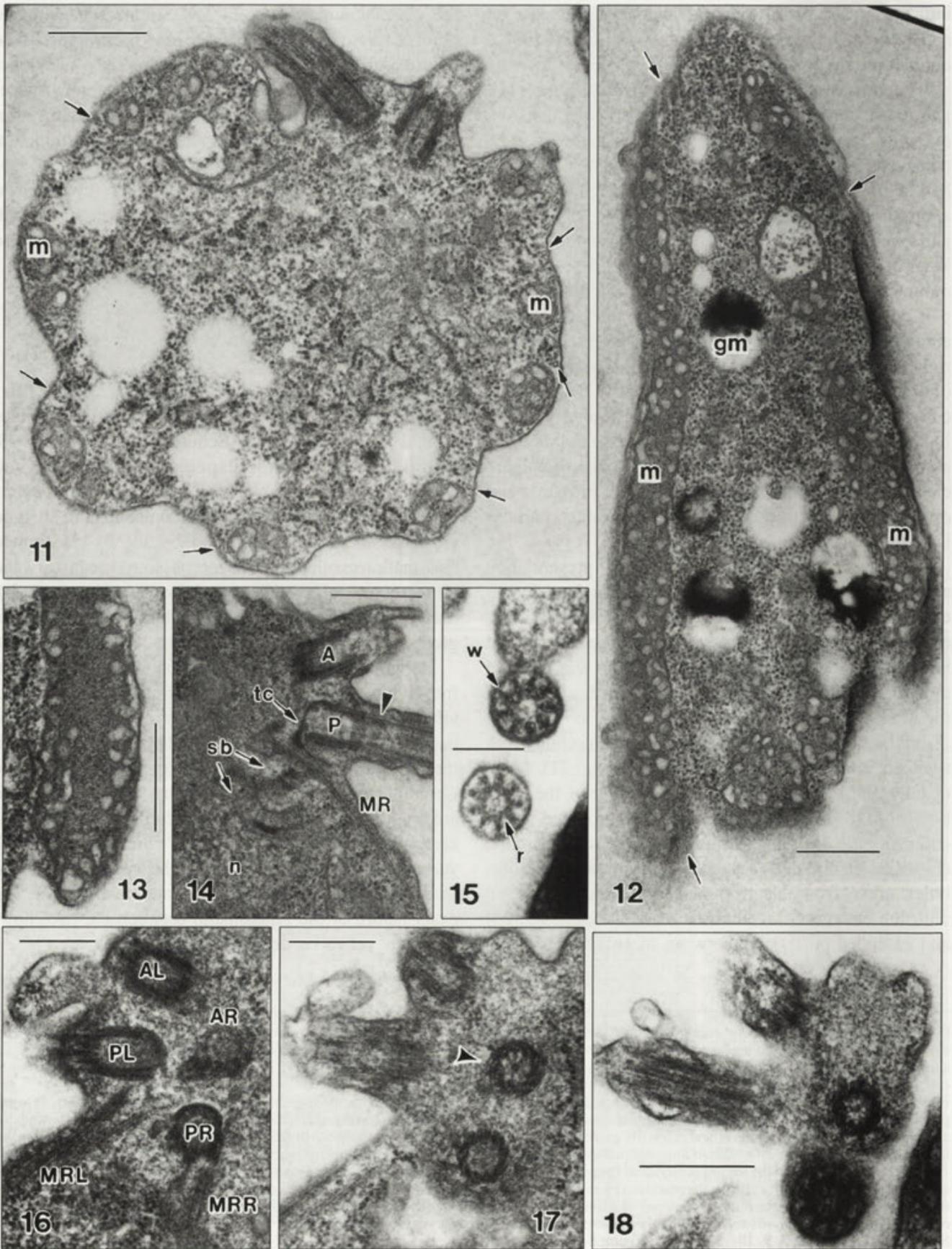
In addition to the root microtubules, cytoskeletal microtubules arose from a focal point near the cell anterior (Fig. 26) and descended posteriorly as a loose corset (Figs. 11, 12, 25).

DISCUSSION

The mode (pseudopodial) and location (cell posterior) of prey capture, the presence of refractile granules around the cell nucleus (Schuster and Pollak 1978, Mylnikov 1987) and, especially, the large and complex paranuclear body, suggest that *Cholamonas cyrtodiopsis* belong to the cercomonads. If so (and the assessment is complicated by the as-yet limited information available on the morphology, life history and ultrastructure of protists that look like cercomonads), then *C. cyrtodiopsis* is a most unusual member of the group.

Previously described cercomonads have all been free-living and all, with the exception of *Cercomonas vibrans* Sandon (Sandon, 1927), have been described as bacterivores. *Cholamonas cyrtodiopsis* is therefore the first gut commensal flagellate to be assigned to the Cercomonadida, and one of the first to make a diet of eukaryotes (perhaps the first recorded mycophagous cercomonad). Since the host insect's diet is one in which yeast are commonly encountered, the association of host insect and endocommensal flagellate makes sense. The exact nature of the relationship between the two, however, has yet to be determined.

Figs. 1-10. *Cholamonas cyrtodiopsis* gen. n., sp. n., light and electron micrographs. 1 - Scanning electron micrograph of a trophic cell, showing the ventral surface with two emergent and two stubby flagella. 2 - Light micrograph (differential interference contrast) of two living cells, showing the positions of nuclei, apical (ga) and median (gm) refractile granules, and food vacuoles (f). 3 - Light micrograph of a protargol-stained cell. The nucleus (n) and flagella, including the stubby flagella (arrow), are visible. Figs. 4-8. Transmission electron micrographs of sectioned cells. 4 - Section through the stubby flagella, showing a reduced axoneme in the more proximally-sectioned flagellum and no axonemal microtubules in the more distally-sectioned flagellum. 5 - Longitudinal section through the apical end of a cell, showing the apical refractile granules (ga) and their positional relationship to the basal bodies of the kinetid, the nucleolus (nu) and the Golgi (g). 6 - Nonmedian longitudinal section in the region posterior to the nucleus, showing the median refractile granules (gm). 7 - Longitudinal section in the region posterior to the nucleus (n), showing a profile of the paranuclear body (p). 8 - Oblique longitudinal section showing an engulfed yeast cell (y), the nucleus (n) and its subtending parabasal body (p), and two median refractile granules (gm). 9 - Light micrograph (differential interference contrast) of a living cell at the moment of yeast cell ingestion. 10 - Light micrograph of protargol-stained cells, showing a *Cholamonas* cell presumably in the process of engulfing a yeast cell (y). Scale bars: 1 - 2 μ m; 2, 3, 9, 10 - 5 μ m; 4 - 300 nm; 5-8 - 1 μ m



Trophic cells of *C. cyrtodiopsis* most closely resemble the swimming stages of species of *Cercomonas* (Schuster and Pollak 1978). However, the gliding locomotion and amoeboid movement found in *Cercomonas* spp. and all other species referred to the cercomonads (except *Discocelis saleuta*), is conspicuously absent from *C. cyrtodiopsis*.

Although *Cholamonas cyrtodiopsis* is biflagellate, as are most species assigned to the Cercomonadida, *C. cyrtodiopsis* lacks the expected anterior flagellum, but instead has two flagella directed posteriorly. Moreover, neither of these two flagella is intimately associated with the ventral cell surface as occurs, for example, in *Cercomonas longicauda* (Mylnikov 1987), *Cercomonas cf. varians* (Mignot and Brugerolle 1975) and *Cercomonas* sp. (Shirkinina 1987).

Each of the two basal body pairs described in the kinetid of *C. cyrtodiopsis* corresponds in many of its features to the single pair found in cercomonads, and especially to those species of *Cercomonas* in which a single, posteriorly-directed microtubular root has been described (Mignot and Brugerolle 1975, Mylnikov 1987). However, if the kinetid of *C. cyrtodiopsis* is derived from a *Cercomonas*-like ancestor, then several major changes have occurred. Firstly, the kinetid has been doubled, and the components of the kinetid have not separated. The phenomenon has not previously been observed among organisms assigned to the Cercomonadida, but similar kinetid doublings are found elsewhere among protists. Both "doubling" and "halving" events occur in green flagellates (Stewart and Mattox 1978, O'Kelly and Floyd 1984); doubling events analogous to what is observed in *Cholamonas cyrtodiopsis* are most evident in the genera *Pyramimonas* e.g. (Moestrup and Hori 1989, Daugbjerg and Moestrup 1992) and *Polytomella* (Brown *et al.* 1976). Doubling events in which the kinetids separate

but remain within a single cell are characteristic of diplomonad flagellates (Brugerolle 1975). Secondly, the anterior flagellum in each pair is a stump, not fully emergent. This phenomenon, fairly common in various protist groups, has not previously been described at the electron-microscope level among organisms referred to cercomonads. Patterson and Zölffel (1991) said of certain genera potentially assignable to *Cercomonas*, "... we presume the original author overlooked a short anterior flagellum". The situation in *C. cyrtodiopsis* raises the possibility that, in some of these doubtful genera, the anterior flagellum may in fact be lacking. Thirdly, the nucleus-enveloping basket of microtubules that emanates from the kinetid in several species of *Cercomonas*, and the single species of *Heteromita* for which there is a reasonable ultrastructural identity, has been lost in *C. cyrtodiopsis*.

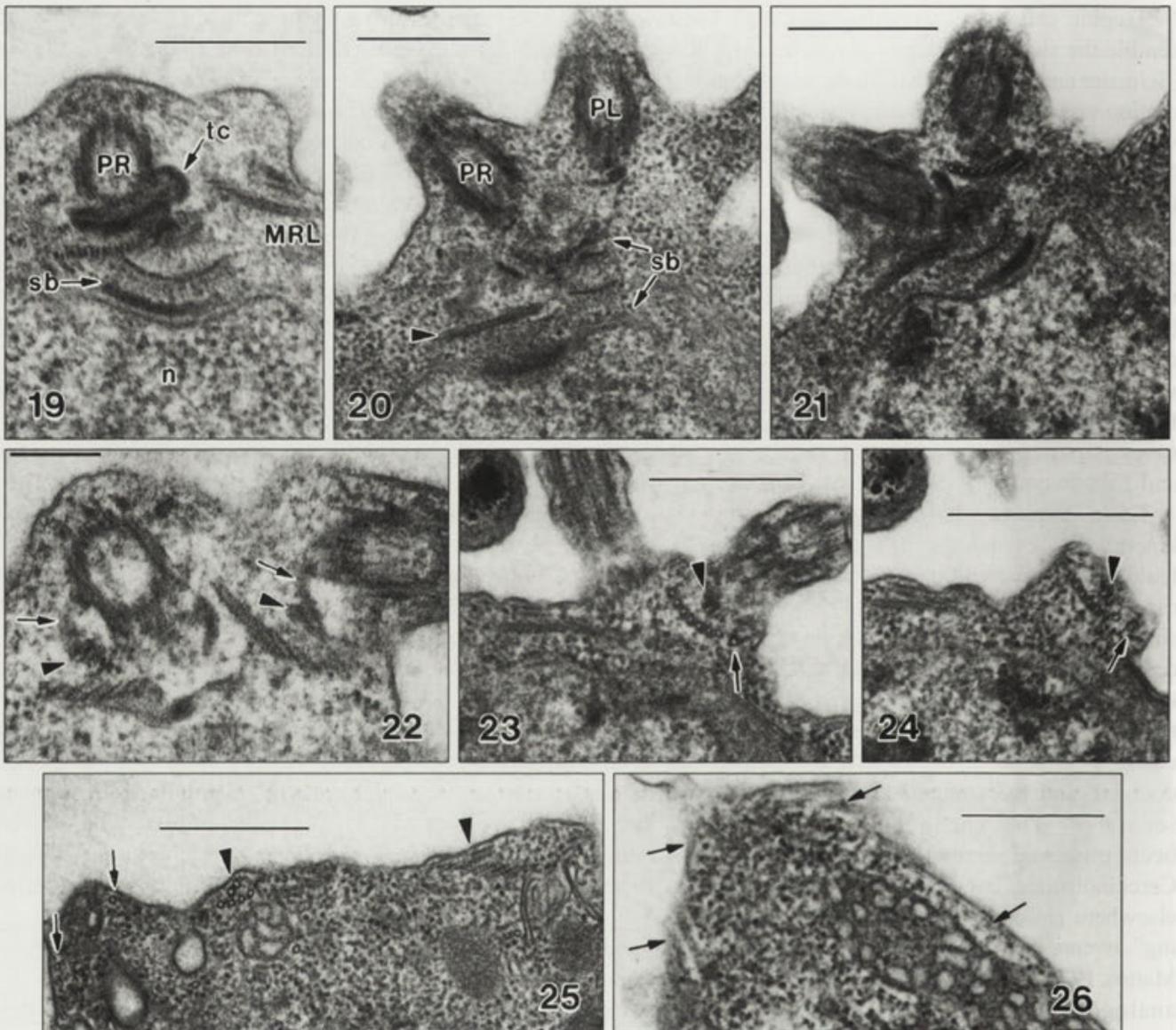
Finally, the apical cap of refractile granules, present in *C. cyrtodiopsis* and apparently consisting of lipid droplets, has no counterpart among described species of cercomonads.

Until more, and more detailed, studies of cercomonads are available, it seems best to treat *C. cyrtodiopsis* as a new genus and species of Cercomonadida. The new species is possibly most closely related to those species of *Cercomonas* that have a single prominent microtubular root, from which *C. cyrtodiopsis* may be derived by doubling of the kinetid, loss of the anterior flagella and perinuclear microtubular basket, and the acquisition of mycophagy and an endocommusal habitat.

DIAGNOSES

***Cholamonas* gen. n.** Cercomonad protists; flagellate trophic cells with longitudinally-arranged peripheral mitochondria; a kinetid having one or more posteriorly

Figs. 11-18. *Cholamonas cyrtodiopsis* - transmission electron micrographs of sectioned cells. **11** - Cross section in the region of the kinetid, showing mitochondrial profiles (m) ranged around the cell periphery. Arrows indicate some of the secondary cytoskeletal microtubules forming a corset around the cell. **12** - Tangential section showing elongated shape of mitochondria, especially at left (m). Some median refractile granules (gm) are also present. Arrows indicate some of the secondary cytoskeletal microtubules forming a corset around the cell. **13** - Mitochondrial profile showing peripheral cristae and crista-free central region. **14** - Section through the kinetid region, showing one anterior (A) basal body bearing a stubby flagellum, one posterior (P) basal body bearing a long flagellum, one of the two microtubular roots (MR), a terminal cap (tc) connecting the posterior basal body to the microtubular root, and a striated band (sb) linking the root to the nucleus (n). The ring structure in the long flagellar transition region is indicated (arrowhead). **15** - Cross section through two flagella in the transition region, one showing the doublet-associated wings (w) in the proximal part of the transition zone, the other showing the ring structure (r) in the distal part of the transition zone. **16-18** - Serial sections showing the absolute configuration of the kinetid, viewed from the dorsal surface of the cell (base-to-tip orientation of basal bodies). **16** - Proximalmost section, showing the left (AL) and right (AR) anterior basal bodies, the left (PL) and right (PR) posterior basal bodies. The left (MRL) and right (MRR) microtubular roots extend from the vicinity of the corresponding posterior basal bodies. Note the offset of the left and right basal body pairs (compare Fig. 1). **17** - Triplet microtubules (arrowhead) are present at the proximal end of the right anterior basal body (compare Fig. 15). **18** - Distalmost section. Scale bars: **11-14, 18** - 500 nm; **15-17** - 300 nm



Figs. 19-26. *Cholamonas cyrtodiopsisidis*, transmission electron micrographs of sectioned cells. 19-25 - Details of kinetid structure, viewed from the cell posterior, ventral side up. 19 - Section near the origin of the right microtubular root near the right posterior basal body (PR), showing the terminal cap (tc) and its association with the basal body and the root. Also present is the prominent striated band (sb) and its attachment to the root as well as to the envelope of the nucleus (n). The left microtubular root (MRL) is also visible. 20 - Section near the origin of the left microtubular root near the left posterior basal body (PL), showing the striated band (sb) in a grazing section (hence it appears less prominent than the bands shown in Figs. 20 and 22). Six microtubules are visible in the root. The right microtubular root (arrowhead) is also visible. 21 - Section of the left microtubular root, slightly posterior to the view in Fig. 20 but from a different cell. Six microtubules are visible. Also present are the right posterior basal body and right microtubular root with its associated striated band. 22 - Origins of the two-stranded components of both the left and right microtubular roots (arrowheads). Both of these two-stranded components are linked to their corresponding basal bodies by a delicate fiber (arrows). 23 - Cross section of the left microtubular root, same cell as Fig. 21 but slightly posterior, near the origins of the two-stranded component (arrowhead) and divergent singlet microtubule (arrow). Seven microtubules are in the main band of the root. The right microtubular root, tangentially sectioned, is visible to the left of the image (as in Figs. 21, 24). 24 - Distal continuation of the series shown in Figs. 21 and 24, showing continuation of the tripartite configuration (arrowhead) of the left microtubular root. 25 - Microtubules are lost from the microtubular roots (arrowheads); the right microtubular root is in cross section). Secondary cytoskeletal microtubules are visible (arrows). 26 - Oblique longitudinal section of a cell near its apex, where secondary cytoskeletal microtubules (arrows) converge. Scale bars: 19- 21, 23- 26 - 500 nm; 22 - 200 nm

directed, compound microtubular roots and lacking a perinuclear microtubular basket; pseudopods not expressed except briefly around the posterior ingestion area at the moment of prey ingestion; prey typically consisting of eukaryotes. Etymology: "Gut flagellate" (*Chola-*, Gk. "intestine"; *monas*, Gk. "wanderer"). A third declension feminine Latin noun. With one species, *C. cyrtodiopsis*.

***Cholamonas cyrtodiopsis* sp. n.** Biflagellate, uninucleate protists in the hindgut of the stalk-eyed fly *Cyrtodiopsis dalmanni*. Cell shape plastic, typically cylindrical with rounded ends, more obovate when compressed; uncompressed cells 5.0 - 8.0 µm long, 2.0 - 5.0 µm wide at the widest point. Refractile granules in two groups, one at the apical end of the cell, the other in the vicinity of the nucleus, the two groups ultrastructurally dissimilar. Kinetid doubled, with two posteriorly directed flagella, two stubby flagella, and two posteriorly directed compound microtubular roots. Feeding on yeast. Type locality: Kuala Lumpur, Malaysia.

Holotype: cryopreserved living material, conserved at the American Type Collection (ATCC) as strain 50325. Protargol-stained slides and resin-embedded cells derived from strain 50325 conserved at ATCC and Natural History Museum, Smithsonian Institution.

Etymology: "from *Cyrtodiopsis*", the genus name of the host stalk-eyed fly.

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