AN ELECTRON MICROSCOPE STUDY OF CORTICAL STRUCTURES OF OPAлина OБTRIGONOIDEA*

BY DOROTHY R. PITELKA, Ph.D.
(From the Department of Zoology, University of California, Berkeley)

PLATES 109 TO 113
(Received for publication, April 13, 1956)

INTRODUCTION

The persistent puzzle of the opalinid protozoa, claimed as ciliates by some authorities and as flagellates by others, intrigues the student of protistan phylogeny and systematics. Any new approach to a study of the cytology of these organisms should help to eliminate some points of dispute and perhaps focus attention on others. An electron microscope study of cortical structures in *Opalina obtrigonoidea* was undertaken toward this end, and also in the hope of extending present knowledge of the organization of the flagellar apparatus in general.

The locomotor organelles of ciliate protozoa are arranged in linear series. These are highly modified in specialized forms, but primitively they are longitudinal rows, or kineties. The basal bodies, or kinetosomes, of such a kinety are usually associated with a longitudinal fiber, or kinetodesma (17). Recent electron microscope studies of three holotrich ciliates, *Paramecium* by Metz, Pitelka, and Westfall (21) and Sedar and Porter (26), *Tetrahymena* by Metz and Westfall (22), and *Colpidium* by Wohlfarth-Bottermann and Pfefferkorn (30), clarify the morphological relationship of cilia, kinetosomes, and kinetodesma in these forms. The kinetodesma is composed of a series of overlapping fibers, each arising from a kinetosome, passing to one side and anteriorly, and ultimately tapering to an end. The kinetodesmal fiber is prominently cross-striated, with repetitions of 300 to 500 Å. The kinetosome itself, seen in Sedar and Porter’s beautiful sections, is a tubular continuation of nine peripheral fibers from the cilium.

Flagella of flagellate protozoa likewise arise from kinetosomes, but the relationships of these with other intracellular bodies vary from one flagellate group to another and may become quite complex. Information concerning their fine structure is exceedingly fragmentary. Recent electron microscope studies of sectioned *Trichomonas muris* by Anderson (1) and *Trypanosoma equiperdum* by Anderson, Saxe, and Beams (2) indicate that axial fibers from the flagellum continue into the kinetosome. In *Trichomonas* the

---

* This investigation was supported by grants (C-2183 and RG-3957) from the Division of Research Grants of the National Institutes of Health, United States Public Health Service.

---

423
latter appears to be a tube of nine fibers generally similar to the kinetosome of *Paramecium*, but its precise limits are not clear.\(^1\)

For a review and bibliography of published reports on opalinids based on light microscope studies, the reader is referred to Grassé (11), who treats the group as a superorder of the Class Zooflagellata. Corliss (7) summarizes arguments concerning classification and agrees that a near-flagellate position is reasonable, while Ulrich (27) gives them separate status equivalent to his Flagellata, Rhizopoda, and so on. Metcalf (19, 20) has been the most active spokesman for a pro-ciliate point of view. A detailed light microscope study of the species considered in the present report was made by Cosgrove (8). One previous electron microscope study of opalinids was the pioneer work of Bretschneider (4) on *O. dimidiata*; his findings are in part confirmed here and will be discussed below.

In this report the locomotor organelles of *Opalina* will be referred to as flagella. This does not indicate that I believe the organisms to be flagellates. There is no consistent morphological distinction between cilia and flagella (25). Descriptively, the term "cilia" usually is used if large numbers of these organelles operate together, but it is also frequently applied by botanists to the relatively few flagella of algal cells, and it is never used for the abundant flagella of hypermastigote flagellates. Simply to avoid the awkward use of both words in every statement referring to both single and multiple organelles, I shall use "flagella" as an inclusive term and "cilia" as an exclusive one applying only to the flagella of true ciliates and of metazoan epithelia.

I am indebted to Miss Jane Westfall for technical assistance, to Dr. Robley Williams for valuable advice concerning sectioning and other techniques, and to Dr. William Balamuth for critical reading of the manuscript. The electron microscope was made available to me through the generosity of Dr. E. A. Steinhaus, Mr. K. M. Hughes, and Dr. A. H. Gold of the Departments of Insect Pathology and of Plant Pathology at the University of California.

**Materials and Methods**

Frogs (*Rana pipiens*) were obtained from commercial sources. The intestine was removed, opened, and washed out with salt solution. Specimens of *Opalina obtrigonoidea* were picked up by pipette and, if necessary, further concentrated by centrifugation.

Pellicular fragments were obtained by two methods. Some samples of *Opalina* were placed in watch glasses and observed under a dissecting binocular while 2 per cent osmium tetroxide was added, a drop at a time, just until all activity ceased. The cells were washed, and then fragmented by sonic vibration in a Raytheon 9-Kc magnetostriction oscillator. The fragments were reconcentrated and were air-dried from microdrops on collodion-coated specimen grids.

Other samples were treated with the detergent, digitonin, utilizing a method developed by Mazia (18) and first applied to protozoa by Child and Mazia (6). Concentrations of fresh *Opalina* were placed in cold (\(-10°C\)) 40 per cent ethanol in centrifuge tubes for 4 hours. At the end of this period the cells had settled to the narrow bottom of the tube and were

\(^1\) An electron microscope study of *Symure* by I. Manton (*Proc. Leeds Phil. Soc.*, 1955, 6, 303), which has just come to my attention, demonstrates a conventional kinetosome of the *Paramecium* type in this chrysomonad flagellate. Also present are slender striated fibers leading from the kinetosome toward the nucleus.

\(^2\) This species was transferred to the genus *Cepedea* by Metcalf (19).
collected with a minimum of fluid in a chilled pipette and transferred in small drops to chilled depression slides containing cold 1 per cent solution of digitonin in distilled water. As the preparation warmed to room temperature the cytoplasm, "solubilized" by the detergent, dispersed, leaving empty pellicular ghosts. The process was watched under a 36 X dissecting binocular and individual ghosts or fragments were isolated by micropipette and washed through six to eight drops of distilled water to free them from cytoplasmic debris. This was usually performed using bright reflected light, since the empty pellicle is so transparent as to be nearly invisible by transmitted light. Pellicle fragments were then pipetted onto coated grids and air-dried. This technique is, at least potentially, less random than sonic fragmentation. Other applications are still in the experimental stage; their success for electron microscopy of protozoan cortical structures seems to depend largely on the presence of a fairly firm pellicle which resists solubilization. Contractile ciliates such as *Stentor* and *Spirostomum* disperse completely, leaving no manageable cortical fragments. The potential importance of this technique for phase microscopy and biochemical studies is discussed by Child and Mazia (6).

Specimens for sectioning were fixed in 1 per cent osmium tetroxide buffered to pH 7.2, dehydrated, embedded in a 9:1 mixture of butyl and methyl methacrylate, and sectioned with a Servall microtome. Thin sections were selected according to their interference colors and mounted by the method of Williams and Kallman (29). They were examined without removing the embedding plastic. The microscope was an RCA EMU-2.

**RESULTS**

The Pellicle.—The flagella of *Opalina* are closely set in longitudinal rows or kineties. Between these kineties the pellicle is formed of numerous ribbon-like bands that parallel them (Figs. 1 and 3). In some fragments, each band can be seen to consist of a number of fine fibers, and in Figs. 6 and 7 these fibers show a periodic structure at spacings of 225 Å. Cross-sections show that the bands lie perpendicular to the cell surface (Fig. 8), forming up to 15, rarely more, continuous longitudinal ribs between adjacent kineties. Anteriorly, where the kineties are more numerous and closer together, there are fewer bands or ribs between them. A delicate membrane bounds the ectoplasm between the ribs and extends to cover the ribs also. The shortest distance separating the ribs in sections presumed to be actually transverse is 360 μ, and the minimum height of the ribs is 610 μ. Bretschneider (4) showed these ribs and described them as pellicular folds. They have also been detected as ridges or crests by many light microscopists (8, 11).

Flagella.—The flagella of *Opalina* exhibit the structure, now thoroughly familiar, characteristic of all flagella investigated (3, 10, 25). Eleven axial fibers may be counted in frayed, digitonin-treated flagella, and micrographs of sections (Fig. 10) show these in the conventional arrangement of nine peripheral fibers around a closely approximated central pair, all embedded in a structureless matrix and bounded by a membrane. Each peripheral fiber is double, each central one single. No evidence has been found of a helical fiber reported by Bretschneider (4). The diameter of the whole flagellum is about 250 μ.

The flagella emerge from the cell surface between the ribs. The cell membrane
about the base of each flagellum forms a conical depression, about 700 m µ in
diameter at its widest, from the bottom of which the flagellum arises. A flagel-
lum torn away from the cell by sonication (Fig. 2) carries a dense girdle around
it at this point, apparently representing the collapsed pellicular receptacle.
The surface membrane of the flagellum is probably continuous with the cell
membrane. At the point where the flagellar axis emerges from the cell there
often, but not always, appears in vertical sections a transverse line (Fig. 14),
which may indicate an incomplete partition across the flagellum at this level,
or may be a reinforced ring in the surface membrane. The two central fibers
of the flagellar axis end here.

Kinetosomes.—Within
the ectoplasm of the cell, the nine peripheral fibers
continue for a distance of slightly less than 400 m µ. The individual fibers are
distinct in fragmented material (Figs. 2 and 6). In sections, they appear to be
partially embedded in a layer of osmiophilic material (Figs. 8 and 14), and
the tube thus formed encloses a substance of very low density similar to the
matrix of the surrounding cytoplasm. The nine fibers all terminate abruptly
without further differentiation. The kinetosome thus is a fibrous cylinder,
open at its inner end and continuous at its outer end with the axis of the flagel-
lum. It constitutes the entire intracellular portion of the flagellum.

Interkinetosomal Bodies.—Alternating
with the kinetosomes in each longi-
tudinal row are small fibrous bodies. In the fragments pictured in Figs. 4 and
6, these interkinetosomal bodies appear nearly rectangular in profile and show
vague striations running parallel to the pellicle surface. Perhaps these longi-
tudinal stripings represent a binding material; they do not appear in other
micrographs. In Fig. 5, the interkinetosomal bodies are disrupted, and numer-
ous fibrils, about 150 A in diameter, are seen, clearly oriented perpendicular
to the cell surface and parallel to the fibers within the kinetosome. These
fibrils are apparent again in sections cut vertically through the interkinetosomal
bodies (Fig. 8).

Where the plane of a tangential section cuts horizontally through a row of
kinetosomes at the level of the interkinetosomal bodies, as in Figs. 10 and 11,
the latter show a distinctly polar arrangement. Each interkinetosomal body
arises from the anterolateral curve of a kinetosome as a pair of strands, each
about 150 A in thickness and separated by an interval of about 300 A. As they
pass anteriorly, the two strands approach and join each other (Fig. 11). In the
micrographs they sometimes are seen to meet the next kinetosome anteriad;
more often they do not. I interpret this to mean that they pass slightly deeper
into the ectoplasm, thus out of the plane of the section in most instances,
before meeting the next kinetosome. Vertical sections (Fig. 8) show them as
continuous with both adjacent kinetosomes in a somewhat zigzag band.

On first inspection it is difficult to reconcile the appearance of the inter-
kinetosomal bodies in Figs. 10 and 11—where they do not look like “bodies”
at all—with their appearance in fragmented material. But careful examination
of Fig. 11 shows in each interkinetosomal strand granules which are cross-sections of the individual fibrils pictured in Fig. 5. The continuity of each strand in Fig. 11 again suggests the presence of an electron-dense matrix which is responsible for cohesion of the constituent fibrils in fragmented preparations. In the latter, the two rows of fibrils, flattened upon one another and probably somewhat shrunken, appear in side view as discrete bodies. The term, "interkinetosomal bodies," is used here for descriptive convenience and should be replaced by a more meaningful one when we are in a better position to understand their significance.

When the asymmetry of interkinetosomal body attachment was first observed, it seemed unlikely that its orientation with respect to body polarity could be determined. However, the section shown in Fig. 10 provided a partial solution to this problem. At the upper left of this figure a row of flagella and neighboring pellicular ribs are cut just above the surface of the ectoplasm. The row of flagella ends above the middle of the picture, and the pellicular ribs clearly converge beyond it, showing that the kinety has truly terminated and not merely passed out of the plane of the section. *Opalina* possesses many incomplete kineties, all originating with accompanying pellicular ribs at the anterolateral margin of the body and ending posteriorly at any level. The figure must represent the posterior terminus of a kinety, since the prolongation of the neighboring kinety and ribs rules out the possibility that this is an anterior section. I still have no means of determining the right-left orientation of this asymmetric pattern.

*Other Intracellular Fibers.*—Bretschneider (4) reported very delicate fibers, 25 m in diameter, running transversely between the flagellar rows just beneath the pellicular ribs in *O. dimidiata.* Unfortunately, his published reproductions do not show these clearly. Cosgrove (8), working with *O. obirigonoidea* stained for light microscopy, reported transverse or oblique fibers connecting the kinetosomes of adjacent rows. Since Bretschneider's fibers were well below the limits of light microscope resolution, Cosgrove's fibers may not represent the same thing. In addition, Cosgrove found thicker (nearly the diameter of the flagella) fibers passing through the cell dorsoventrally. These arose from kinetosomes and sometimes could be followed to kinetosomes of the opposite surface. They were sinuous, rarely if ever following straight courses. Other workers have reported transverse fibers or endoplasmic fiber lattices in various opalinids (13, 11).

In my fragmented material there obviously are no fibers other than those already noted. In tangential sections, immediately beneath the cell membrane, at the bases of the pellicular ribs, minute circular, elliptical or rod-shaped profiles occur in abundance, rather regularly arranged in oblique rows (Figs. 10–12). In diameter the profiles vary from 25 m or less to 100 m. In addition one sees, rarely, a very fine fiber, estimated at 15 to 20 m in diameter, accompanying such a row. The rows are spaced at intervals slightly
less than the longitudinal distance between kinetosomes and show no morphological relationship to them in my material. No endoplasmic fibers of any sort are detectable.

Falx.—Efforts were made throughout this study to identify in electron micrographs the falx, described by Cosgrove (8) as a double (single more posteriorly) row of heavy flagella near the anterolateral margin of the body, underlain by heavy falcular fibers in the ectoplasm. This structure is recognized by Chatton and Brachon (5) as the primary kinety of opalinids. All other flagellar rows originate from the falx and presumably elongate posteriorly by kinetosomal multiplication. Okajima (23), in an experimental study on the metachronal wave in Opalina, found this region to be the most excitable part of the cell surface.

If the falx is represented at all in my material it has no intrinsic morphological features differentiating it from other kineties. Circumstantial evidence suggests that Fig. 8 may represent a section through the falx at the level where it is a single row. Here three rows of flagella occur very close together, separated by only two or three pellicular ribs. Such an arrangement would be expected in the vicinity of the falx, where longitudinal somatic rows join it as illustrated diagrammatically in Fig. 9, or possibly at the posterior pole of the cell, where a reduced number of kineties converges toward a small naked area. In addition, the pellicular ribs and flagella in Fig. 8 are sectioned at rather different angles on the two sides of the presumed falcular area; this also would be expected (see Fig. 9), but would not be so pronounced at the posterior pole. This sort of arrangement occurs with sufficient frequency (15 per cent) in randomly selected micrographs of cortical sections to convince me that most of them could not represent posterior sections. The posterior region of convergence involves a very small area of body surface, while the falx extends for about one-third the length of the cell.

If this interpretation is correct, the kinetosome marked by the arrow in Fig. 8 may represent one of those of the falx; on either side of it are proximal sections of somatic flagellar rows. The supposed falcular kinetosome perhaps is heavier than comparable sections of other kineties, but not markedly so.

Cytoplasmic Bodies.—Spherules of varying size and distribution in the ectoplasm and endoplasm of opalinids have been described by many authors and their significance has been widely debated. Hunter (14) found that “small” granules in both cytoplasmic regions of O. carolinensis responded positively to tests for mitochondria, while “large” granules in both regions were amino acid-containing bodies. Bretschneider (4) distinguished, in electron micrographs of O. dimidia, ectosomes, 160 to 400 μm in diameter, and endosomes, 500 μm to 2 μ. He stated, in agreement with some other workers, that their abundance, position, and structure varied with the age and nutritional condition of the organism.

In my preparations of O. obtigonoidea there is no clear demarcation between
ectoplasm and endoplasm. The number and appearance of protoplasmic bodies vary within wide extremes. Some of the host frogs used had been kept in a cold room without food for considerable periods before examination, hence the nutritional history of the opalinids was complicated by this as well as normally variable factors. In general three types of cytoplasmic bodies, not always mutually distinguishable, appear in my micrographs (Figs. 8 and 13). Briefly they may be described as (1) a small (average 600 m/z long diameter) body occurring in a region within about 1 to 2 m under the surface and containing an irregular scattering of granular or fibrous matter; (2) a larger (average 2.6 m long diameter) body variably abundant throughout the endoplasm and characterized by a peripheral concentration of electron-dense material; and (3) a still larger (2.5 to 4 m) body appearing in some individuals regularly aligned near the cell periphery, and rather uniformly filled with fine granular material. All three are clearly defined by membranes. Their uniformly elliptical shape may be an effect of compression during sectioning (29). Nuclei, when present in the section, appear as in Fig. 8 and are 3.5 to 5 m in diameter. Further clarification of the structure of these various bodies, or attempts to correlate them with granules described from light microscopy, are beyond the scope of this paper.

**DISCUSSION**

The fine structure of the pellicle seen here is, as far as I know, unique. It may be characteristic of all opalinids, as pellicular crests have been described in other genera by light microscopists (11). The body of *Opalina* is flexible and elastic under mechanical distortion, but is not known to undergo spontaneous changes of shape. The pellicular ribs, lying as they do perpendicular to the surface, seem to provide a happy solution to the need for flexible mechanical support, without covering the surface with a continuous layer of skeletal material, and this conceivably may be of some significance for an organism that, according to majority opinion, feeds saprozoically.

Flagellum structure in the opalinids is conventional, as noted above, and needs no discussion. Their kinetosomes are markedly similar in structure to those of *Paramecium*, studied in section by Sedar and Porter (26), and apparently also to those of *Trichomonas* (1). These all differ to some extent from the kinetosomes of metazoan cilia studied by Fawcett and Porter (10) and Bradfield (3). Features of the latter that have not been found in protistan kinetosomes are a denser osmiophilic shell, always with a tapering, closed, inner end; prominent lateral bulges or curvatures (lacking in some types) that give the kinetosome a pronounced polarity, supposedly coincident with plane of ciliary beat; and root fibers (poorly developed or lacking in mammalian epithelia studied) with a complex pattern of cross-striations, extending vertically into the cell.

Instead of vertical root fibers, ciliate kinetosomes bear anteriorly directed
kinetodesmal fibers. The interkinetosomal bodies of *Opalina* are obviously similar to these in general position. But they do not overlap to form a continuous kinetodesma, and in structural detail they appear rather different. Instead of being solid and uniformly cross-striated, they are compounded of discrete subfibrils, and these attach to the kinetosome in two separate rows that merge anteriorly and then probably meet the next anterior kinetosome.

The validity of this comparison of opalinid with ciliate structures may be seriously limited by the fact that well documented investigations of the ultrastructure of kinetal organelles in ciliates are presently confined to three genera of hymenostome holotrichs, and we do not know to what extent simpler, or more complex, ciliates are similar to these. A preliminary, unillustrated report by Fauré-Fremiet and Rouiller (9) on sectioned *Stentor* suggests that kinetodesmata here may be quite unlike those of hymenostomes.

Comparisons with flagellates are still more difficult, since no single species has been studied as thoroughly as *Paramecium*. Kinetosomes of *Trichomonas* seem to be similar to those of *Paramecium*, but the interesting micrographs of a trypanosome by Anderson, Saxe, and Beams (2) and of a chrysomonad by Wolken and Palade (31) do not include enough information on kinetosomes to justify any conclusions in this connection.

There is little reason to suppose that we can, for some time to come, do much to clarify phylogenetic relationships of opalinids on the basis of structure of the flagellar apparatus; the answer to this question may come rather from studies of life history and morphogenesis. But as morphological information on additional ciliates and flagellates accumulates, the nature of opalinid structures may be peculiarly significant for the consideration of a much broader question: whether and to what extent the physiological mechanisms of flagellar movement and coordination require in the cortex of all flagellated cells a similar morphological framework, regardless of degree of evolutionary kinship.

Ulrich (27) cogently remarks that efforts to classify the opalinids have been too much influenced by the assumption that ciliated cells must differ in some fundamental way from other flagellated cells. This assumption is understandable, considering the obvious requirement for coordinating mechanisms in ciliated cells, but the very fact of this requirement should warn us against ascribing taxonomic significance to it. If there are constant morphological differences, we should inquire first whether they are related to functional differences.

There is no consistent morphological distinction between cilia and other flagella in their extracellular parts (25). Kinetosomes in all flagellated cells are closely associated with fibrous protein structures and may be important in their morphogenesis; in addition they probably initiate or translate the immediate impulse for flagellum contraction. If we examine the admittedly inadequate array of examples whose fine structure has been revealed, the
most impressive distinction separates kinetosomes of metazoan ciliated epithelia from those of protists.

Actually, this distinction may be a functionally significant one. In metazoan ciliated epithelia, according to Gray (12) and Párducz (24), each cilium executes its entire beat within a single, inherently fixed plane. The direction of the effective stroke is in most instances irreversible, as is also the direction of passage of metachronal (coordinating) waves. Opalinids and ciliates, on the other hand, are able to change readily and within wide limits both the plane of beat of individual flagella and also the direction of propagation of metachronal waves (23, 24). In addition, each flagellum during its recovery stroke bends more or less sharply out of the plane of its effective beat (24). In flagellates, in which one or a few flagella are present, the organelle is capable of the widest variety of contortions, some of which have been analyzed by Lowndes (15, 16). Thus protistan flagella, including cilia, are specialized for versatility, those of metazoa for constancy.

The marked polarity of most metazoan kinetosomes may reflect this fact, but the equally marked polarity of ciliate kinetodesmata and opalinid interkinetosomal bodies must be related to the morphological and physiological polarity of the whole cell, rather than to the movement of individual flagella. The kinetodesmata of ciliates are undoubtedly important in morphogenesis (17) but their significance in metachronal coordination has not been conclusively demonstrated (26, 28).

**SUMMARY**

The pellicular framework of *Opalina obtrigonoidea* consists of numerous longitudinal ribs parallel to the kineties. These ribs lie erect on the cell surface, and each is composed of striated longitudinal fibers. A membrane covers the ribs and the ectoplasm between them. Flagella, of conventional structure, emerge from the ectoplasm between the ribs. The two central fibers of each flagellum end at the cell surface; the nine peripheral fibers continue for about 400 μm into the cell to form an open tubular kinetosome. From the anterolateral curvature of each kinetosome arise two rows of fibrils, each fibril oriented perpendicular to the cell surface and about 150 A in diameter. The two rows converge anteriorly and probably meet the next adjacent kinetosome. Minute granules or tubules, arranged in oblique rows and at least sometimes accompanied by very fine fibers, lie at the surface of the ectoplasm but show no detectable connection with the kinetosomes. The whole flagellar apparatus of *Opalina* thus bears a general resemblance to the infraciliature of some holotrich ciliates, but the degree of evolutionary relationship between them remains uncertain.

**BIBLIOGRAPHY**

REFERENCES

CORTICAL STRUCTURES OF OPALINA OBTRIGONOIDEA


EXPLANATION OF PLATES

PLATE 109

Fig. 1. Low power micrograph of a fragment of the pellicle of *Opalina* prepared by digitonin treatment. The flagella arise in orderly rows separated by regions that here show longitudinal striping. Shadowed with palladium. × 3,400.

Fig. 2. Flagellum isolated from cell by sonic fragmentation. Longitudinal fibers of flagellum and of kinetosome are seen. Dense girdle around flagellum just above kinetosome is collapsed pellicular receptacle. Shadowed with palladium. × 22,400.

Fig. 3. Higher power view of a small area of digitonin-treated pellicle, seen from the outside. Kinetosomes are seen in outline through the pellicle, which is composed of fibrous bands overlapping laterally. Shadowed with palladium. × 17,600.
Pitelka: Cortical structures of *Opalina obirigonoidea*
Fig. 4. Part of a kinety isolated with two neighboring pellicular bands by digitonin treatment. Kinetosomes and interkinetosomal bodies are seen in profile. Shadowed with palladium. × 17,300.

Fig. 5. Part of kinety isolated by sonic fragmentation. Flagella have broken off and interkinetosomal bodies are partially disrupted, exposing fibers of a maximum length of about 300 μm, and about 150 Å in diameter. These are arranged parallel to the fibers of the kinetosome. Shadowed with palladium. × 30,400.

Fig. 6. Part of kinety and adjacent pellicular bands isolated by sonic fragmentation. Flagella are broken off. Note three-dimensional effect of open fibrous tube at bases of kinetosomes. Shadowed with palladium. × 17,800.

Fig. 7. Enlargement of part of Fig. 6, showing striated fibers in frayed pellicular band. Period is about 225 Å. × 30,800.
(Pitelka: Cortical structures of *Opalina oblongoides*)
PLATE 111

Fig. 8. Section cut almost vertically into the cell. Sectioned kineties are numbered 1 to 7 clockwise. Pellicular ribs are shown, erect on the cell surface; the fibrous nature of these is seen most clearly at the top of the micrograph. The orientation of ribs and kinetosomes changes abruptly at kinety 4, which is separated from kinety 5 by a single, (perhaps two) obliquely cut, pellicular rib. Arrow marks kinetosome of kinety 4, which may be one of those of the falx. Several interkinetosomal bodies along a segment of kinety 5 are cut almost longitudinally. Each appears to be continuous with adjacent kinetosomes on both sides, the whole forming a somewhat zigzag band.

F, flagellum; KS, kinetosome; IKB, interkinetosomal substance; PR, pellicular rib; CM, cell membrane; T1, type 1 cytoplasmic body; T2, type 2 cytoplasmic body; N, nucleus. \( \times 14,000 \).

Fig. 9. Diagram of anterolateral surface of cell body, drawn from light microscope study and from descriptions and illustrations by Cosgrove (8). Falx and somatic kineties represented as solid lines. Note heavy paired falcular fibers merging posteriorly, and somatic dorsal and ventral kineties diverging from falx. Pellicular ribs, parallel to somatic kineties, are not shown.
PLATE 112

Fig. 10. Section cut tangentially through one kinety at the level of the kinetosomes and through the posterior end of another at the level of the pellicular ribs. The polarity and asymmetry of interkinetosomal body attachment are conspicuous. × 19,000.

Fig. 11. Enlargement of part of Fig. 10. The origin of the interkinetosomal body as two separate strands is clear; each strand seems to attach to one kinetosomal fiber (arrow). Note the orientation of minute granules, circles, and double rods in regular rows above the kinetosomes, but independent of them. These rows include occasional wispy oblique fibers. × 29,600.
(Pitelka: Cortical structures of Opalina obtrigonoidea)
PLATE 113

Fig. 12. Thick tangential section cutting through the pellicular ribs and the surface of the ectoplasm, showing the orientation of granules, circles, and rods in oblique rows. × 18,800.

Fig. 13. Vertical section showing the regular alignment of type 3 cytoplasmic bodies near the cell periphery. × 8,100.

Fig. 14. Longitudinal section through three adjacent flagella at their bases. The middle one shows that the central axial fibers end at the cell surface. A rather sharp line across the flagellum at this point appears to be continuous with the cell membrane and may be a reinforced ring in it at this level, or may be a transverse partition in the flagellum. The kinetosomal fibers appear slightly thickened within the cell. × 24,600.
(Pitelka: Cortical structures of *Opalina obtrigonoides*)