Studies on *Conchophthirius mytili* De Morgan.

I. Morphology and division.

By

George W. Kidder.

(With 5 figures in the text and plate 1—4.)

Contents.

Introduction.

EHRENBERG (1838) described a parasite from the mantle cavity of the fresh water molluscs *Anodonta* and *Unio* under the name of *Leucophrys anodonta*. STEIN (1861) removed this form from the genus *Leucophrys* and created the genus *Conchophthirius* for its reception. He described a second species, *C. steenstrupii*, from *Succinea amphilia*. Since that time ENGELMANN (1862) added the species *curtus* to the genus and DE MORGAN (1925) gave the name *Conchophthirius mytili* to a form described by him from *Mytilus edulus*. In reviewing the literature on the genus, DE MORGAN recognizes the species: *C. anodontae* (EHRENBERG) STEIN, from *Anodonta* and *Unio*; *C. steenstrupii* STEIN from *Succinea*; *C. curtus* ENGELMANN from *Anodon*; *C. mytili* DE MORGAN from the "common salt water mussel". To this list should be added *C. antedonis* ANDRE (1910) from the intestine of certain Crinoidea.

DE MORGAN (1925), in his description of *Conchophthirius mytili*, gave only a sketchy and imperfect picture of that form. Most of his observations were made on living ciliates.

Because of the large size, common occurrence and interesting structure it was thought advisable to investigate in detail this inhabitant of the salt water mussel. This paper deals with the general morphology, cytology and nuclear and cytoplasmic phenomena during division.

The study reported here was carried on at the Zoological Laboratory of Columbia University in New York City and at the Marine Biological Laboratory at Woods Hole, Massachusetts. I wish to acknowledge the helpful suggestions and criticisms of Prof. GARY N. CALKINS throughout this investigation. I wish also to express my appreciation to Prof. FRANZ SCHRADER for helpful suggestions and a friendly interest during the progress of the work.

Material.

Conchophthirius mytili is found creeping about over the muscles and foot of the common mussel, *Mytilus edulus*. Mussels were collected from a variety of localities about New York. Many of the collections proved sterile or so lightly infected as to be impractical. The heaviest infections were found in the mussels gathered at low tide from the rock breakwaters at Brighton Beach on Coney Island. Manhattan Beach mussels showed a lighter infection. Other collections were made at Port Washington and Pelham Bay on Long Island Sound.

The collections of mussels from the pilings of the dock at the Bureau of Fisheries at Woods Hole, Massachusetts, proved the most fruitful. Infection was much heavier per mussel and practically all the Mytili were found to contain the ciliates.

The mussels were collected in a thermos jug and brought as quickly as possible to the laboratory. They were transferred to glass aquaria supplied with running sea water (Woods Hole) or to glass aquaria supplied with a jet of air (New York). In the former situation the ciliates lived very well for weeks but I was able to keep them alive for only a few days in the New York aquaria, although the concentration of the water was kept nearly constant. In both cases the mussels themselves remained alive and vigorous for months. This extreme sensitivity to environmental conditions was also noted in the short length of time the ciliates would live when removed from the host. One to two hours was the limit of their activity, cytolysis taking place rapidly at death.

Many attempts were made to prepare a medium in which the ciliates would live but all have failed.

The infection is never heavy, fifty to seventy-five organisms per mussel being a good find, while the average is five to ten ciliates. In general the larger mussels yield the greater number of forms.

Conchophthirius mytili is, no doubt, to be looked upon as a commensal. Its location in the host and the fact that its food vacuoles are loaded with algae argue against a parasitic nature.

Methods.

The mussels were opened by slipping a scalpel between the valves and cutting the adductor muscle. The water held within the mantle cavity was drained into a syracuse watch glass and with a pipette this water was forced over the foot and muscles several times, washing the ciliates into the dish. With the aid of a dissecting microscope the ciliates were picked out of the water with a fine pipette, as one would do when dealing with free-living forms.

A variety of fixatives were used. SCHAUDINN'S, sublimate acetic in 95 % alcohol, BOUIN'S, ZENKER'S, GILSON-CARNOY'S and strong FLEMMING'S fluids gave uniformly good results when the nuclear elements were to be stained. BENDA'S and CHAMPY'S fluids were used for the preservation of cytoplasmic elements.

The general stains used were HEIDENHAIN'S and DELAFIELD'S haematoxylins, the BORREL stain and the FEULGEN nuclear reaction.

For the study of the neuromotor apparatus whole mounts were stained in HEIDENHAIN'S haematoxylin after fixation in FLEMMING'S fluid, and MALLORY'S triple stain was employed after ZENKER'S fluid fixation. The concentrations and times of the latter technique were those employed by SHARP (1914) and YOCOM (1918).

I should like to insert at this point a remark that may prove useful to those working on forms that are difficult to destain after haematoxylin. For fine differentiation of the neuromotor elements in whole mounts excellent results were obtained by staining in warm (50° C.) iron alum haematoxylin and destaining in a 10 % solution of commercial hydrogen peroxide.

Material to be sectioned was fixed in ZENKER'S, strong FLEMMING'S and BOUIN'S fluids by spurting the ciliates into a dish containing the fixatives. They were then washed, dehydrated and tinted with eosin. Lefevre dishes were found very useful in the dehydration, the ciliates remaining in the groove of the dish as the various liquids were drawn off. From xylol the ciliates were spurted onto the flat surface of a paraffin block. The xylol dissolved a layer of paraffin and the organisms sank into this mixture. They were allowed to impregnate for thirty minutes. The individual ciliates were then picked up with the tip of a small scalpel and embedded in a button of paraffin. They were then oriented and cut at various thicknesses. HEIDENHAIN'S haematoxylin and MALLORY'S triple stains proved most helpful for neuromotor parts on sectioned material.

Many attempts to demonstrate a "silver-line" system were made, using the method of KLEIN (1926) and numerous modifications. *Conchophthirius mytili* always gave negative results, becoming a muddy yellow. On the same slides were numerous *Ancistrum mytili* in which a beautiful "siver-line" system invariably appeared.

For a demonstration of the mitochondria the methods of BENDA (alizarin-crystal violet) and of BENSLEY (acid fuchsin methyl green) gave excellent results.

Special techniques used for the demonstration of the GOLGI elements were KOLACHEV, WEIGL and the modifications of HIRSCHLER. These were the least satisfactory of the techniques used on the organism.

Neutral red (GRUEBLER, and COLEMAN and BELL) and JANUS green B (HOESCHT and GRUEBLER) were used as vital stains.

Observations on living material.

Shape and size.

Orientation, for descriptive purposes, is not uniform in the literature as regards *Conchophthirius*. DE MORGAN (1925) regards the slightly concave surface that is in contact with the substrate as the ventral surface while the mouth is described as lateral. REICHENOW (1929), however, regards the oral surface as ventral and describes the members of the genus as being strongly flattened laterally, creeping about on their left surfaces. Moreover, DE MORGAN regards as anterior the end nearest which the mouth is located. If this latter view is to be followed then the ciliate must be described as continually moving in a posterior direction. While recognizing the oral surface as the physiological ventral surface, for purposes of clearness in the description I shall regard the morphological ventral surface as that which is in contact with the substrate, and the anterior end as the end directed in advance in motion, either creeping or swimming.

In outline *Conchophthirius mytili* roughly resembles a lima bean. It is an irregular oval, flattened dorso-ventrally, slightly narrower at its anterior end and weakly truncate posteriorly (Pl. 1 Figs. 1, 2, 3). The average measurements of one hundred living organisms taken at random were 170.8 μ in length by 108.5 μ in width. The greatest length was 202 μ while the greatest width was 161 μ. The smallest length was 132 μ and the smallest width was 76 μ. These measurements were taken with the aid of a filar micrometer on organisms that were free in the water. If a cover glass is placed over the ciliate it is flattened down so that the width is proportionately greater. Fixation causes shrinkage so that measurements of fixed specimens can give only relative values. As a check on the amount of shrinkage produced single organisms were measured while alive, then fixed and measured. Fixation in SCHAUDINN'S fluid, sublimate-acetic in 95 % alcohol and GILSON-CARNOY'S caused a shrinkage of approximately 20 %. BOUIN'S caused a shrinkage of 17 %, ZENKER'S one of 11 %, CHAMPY'S fluid one of 2 % while osmic vapor caused so slight a shrinkage that it could not be measured.

About one-third of the distance back on the right lateral margin an abrupt dip marks the beginning of the peristomal groove. This groove extends nearly to the posterior of the organism, starting as a narrow depression and ending in a relatively wide fossa which marks the mouth opening. The floor of the groove is pouched out

into a shelf, rising gradually from the anterior end of the groove and being cut off abruptly at the opening of the mouth (Text-Fig. 1, *sh*). From this opening a shallow cytopharynx narrowing into a long gullet can be seen to extend into the endoplasm in a slightly antero-dorsal direction, terminating about two-thirds of the distance across the body.

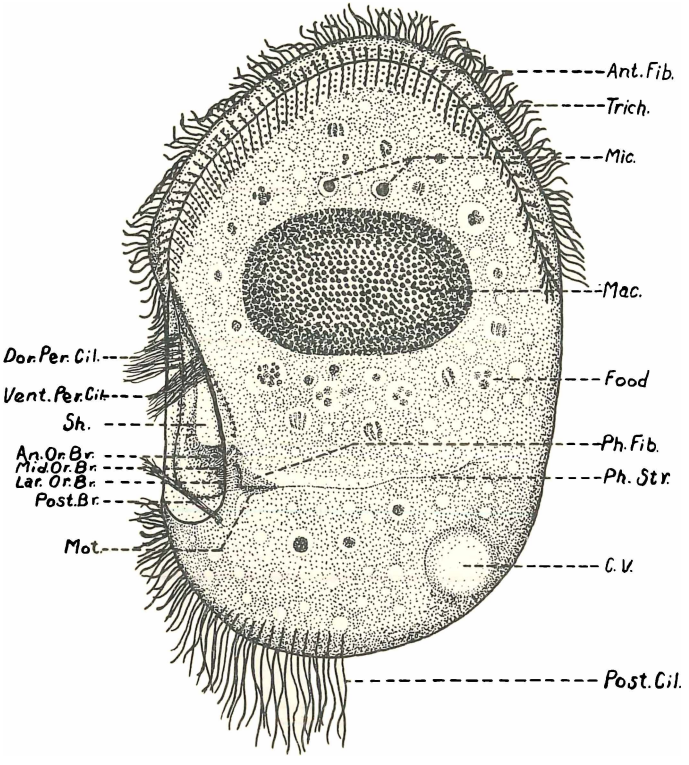


Fig. 1. Diagrammatic figure of *Conchophthirius mytili* from the ventral side. Most of the cilia have been omitted for the sake of clearness. The longitudinal ciliary fibers have been omitted except at their origin in the anterior fiber. × 500. Abbreviations: *an. or. br.*, anterior oral brush; *ant. fib.*, anterior fiber; *c. v.*, contractile vacuole; *dor. per. cil.*, a portion of the dorsal peristomal ciliary row; *food*, food vacuoles; *lar. or, br.*, large oral brush just ventral to the mouth; *mac.*, macronucleus; *mic.*, micronuclei; *mid. or. br.*, middle oral brush; *mot.* motorium; *ph. fib.*, pharyngeal fibers; *ph. st.*, pharyngeal strand; *post. br.*, immotile posterior brush; *post. cil.*, long posterior cilia; *sh.*, shelf; *trich.*, position of trichocysts; *vent. per. cil.*, a portion of the ventral peristomal ciliary row.

The dorsal surface of the organism is slightly convex while the ventral surface is nearly flat in the anterior half, becoming slightly concave posteriorly.

Normally the ciliates move about in a jerky manner adhering closely to the muscle surface with their ventral sides in contact. Occasionally, after they have been washed into a syracuse dish, they will swim freely, rotating on a diagonal axis. These flights are usually confined to the distance between the bottom of the dish and the surface film of the water. They have great powers of adhesion which make it very difficult to draw them up in a pipette. In fact, so firmly do they cling to the bottom of the dish, it is usually necessary to dislodge them by the force of a stream of water from the pipette.

The ectoplasm, as seen in life, is a rather thin, clear area just under the pellicle. It is seen to be very finely granular as compared to the more densely granular endoplasm. The pellicle is very thick but quite elastic. It is thrown into longitudinal folds which traverse the body of the ciliate corresponding to the ciliary lines.

Peripheral cilia.

The body is covered with closely set parallel rows of rather fine cilia. The cilia are more sparsely placed in the posterior regions and are somewhat longer and less active than those of the anterior portion of the body. All of the cilia are directed nearly backward in life and under low powers of the microscope they can be seen to be beating metachronously. This gives the impression of a series of waves originating at the anterior end, to the right of the center, and proceeding diagonally across the organism to the posterior left side. Examination of the organism under the oil immersion lens shows the cilia to orginate beneath the pellicle in rather large basal bodies. These bodies lie beneath the trenches between the longitudinal pellicular ridges (Text-Fig. 3, *b. b.* Pl. 1 Fig. 4).

Peristomal and oral cilia.

Two rows of long, fine cilia originate within the peristomal groove near its anterior end. Under low magnifications these appear as separate undulating membranes, but can be seen to be composed of unfused cilia when viewed under the higher powers of the microscope. These rows of cilia proceed posteriorly, one following the lowest depression of the groove, ventral to the shelf (Text-Fig. 1, *vent. per. cil.*). This ends just anterior to the mouth opening. The other row follows a corresponding depression dorsal to the shelf and swings down in a wide arc in a ventral direction toward the mouth opening (Text-Fig. 1, *dor. per. cil.*). These two rows of cilia are in

constant motion creating a steady current converging at the anterior tip of the peristomal groove and sweeping back toward the mouth.

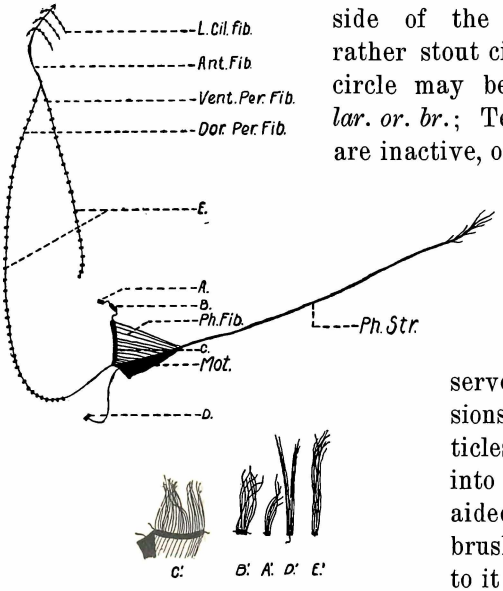


Fig. 2. Diagram showing the parts of the neuromotor apparatus as viewed from the ventral side. Abbreviations: *A.*, basal plate of anterior oral brush; *A'.*, same with cilia; *ant., fib.*, anterior fiber; *B.*, basal plate of middle oral brush; *B'.*, same with cilia; *C.*, basal plate of large oral brush; *C'.*, same with cilia and showing its connection with the motorium; *D.*, basal plate of the posterior brush; *D'.*, same with cilia; *dor. per. fib.*, fiber of dorsal peristomal ciliary row; *E.*, basal plates of the dorsal and ventral peristomal cilia; *E'.*, same with cilia; *l. cil. fib.*, longitudinal ciliary fibers; *mot.*, motorium; *ph. fib.*, pharyngeal fibers; *ph. st.*, pharyngeal strand; *vent. per. fib.*, fiber of ventral peristomal ciliary row.

Surrounding the ventral posterior side of the mouth a wide brush of rather stout cilia in the form of a semicircle may be seen in life (Text-Fig. 1, *lar. or. br.*; Text-Fig. 2, *c'*). These cilia are inactive, or nearly so, until a particle of solid matter has been swept into the lower end of the groove. Then they start a rhythmic beating, sweeping as one up against the particle. I have observed this on numerous occasions and have seen the particles pushed, little by little, into the gullet. This brush is aided by two much smaller brushes located a little anterior to it (Text-Fig. 1, *an. or. br.* and *mid. or. br.*; Text-Fig. 2, *a'*, *b'*). They beat in a posterior dorsal direction against the particles. This curious timing of the beats seems to be entirely independent of the movements of the rest of the cilia throughout the body.

Just posterior to the arc of the dorsal row of peristomal cilia is found another tuft of stout cilia (Text-Fig. 1, *post. br.*; Text-Fig. 2, *D'*). These cilia point outward in a slightly anterior direction. They seem to be quite stationary. I have watched them for long periods of time and have never seen them beat although they may be bent readily by the movements of the organism. I am at a loss to account for their function.

Trichocysts.

In the original description of *Conchophthirius mytili*, De Morgan (1925) states that there appear to be no trichocysts. I find them in abundance, and although they are relatively small and exhibit peculiarities, they can be seen easily, even in the living organism under high magnifications.

These trichocysts have the form of small sacs and are found suspended from the pellicular ridges. They extend through the ectoplasm (Text-Fig. 1 and 3, *trich.* Pl. 1 Fig. 4). They are found in greater numbers on the ventral surface than on the dorsal. I have never succeeded in bringing about their discharge with dilute

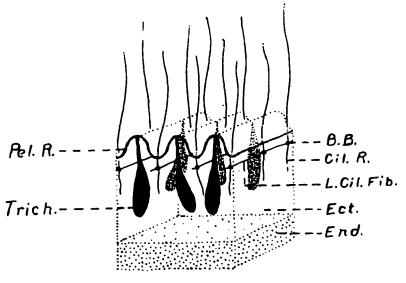


Fig. 3. Schema of ectoplasmics tructure. Abbreviations: *b. b.*, basal body; *cil. r.*, ciliary rootlet; *ect.*, ectoplasm; *end.*, endoplasm; *l. cil. fib.*, longitudinal ciliary fiber; *pel. r.*, pellicular ridge; *trich.*, trichocyst.

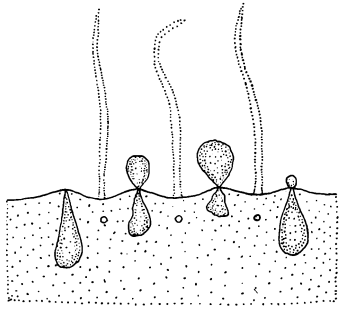


Fig. 4. Optical section of a portion of a flattened organism stained intra vitam with neutral red showing the squeezing out of the fluid contents of the trichocysts.

acids. Lynch (1929) reports seemingly static trichocysts in *Entorhipidium echini* from the sea urchin *Strongylocentrotus*. He was never able to cause their discharge.

A very peculiar property of the trichocysts of *Conchophthirius mytili* is their affinity for neutral red, even in very weak concentration. In fact the trichocysts are the only components of the body that do stain in life with this dye. By this method they are brought out in sharp contrast to the colorless cytoplasm. As the water evaporates and the ciliate becomes flattened the content of each trichocyst sac, stained a bright cherry red, is seen to be squeezed out through the pellicle and hang as a drop on the surface. This drop enlarges until the whole trichocyst disappears, and then bursts like a bubble and vanishes. Text-Fig. 4 gives a diagrammatic representation of this phenomenon. Whether these trichocysts, composed of fluid-filled bags, are functional or not I can not say.

Nuclei

The macronucleus of *Conchophthirius mytili* is an extremely large oval, averaging 50 μ in length and 35 μ in width. It occupies a mid position in the anterior third of the body (Pl. 1 Fig. 1). It is very conspicuous in life as a clear, granular body, usually surrounded by food inclusions. Rarely are food vacuoles seen either dorsal or ventral to the macronucleus. Cross sections of the ciliate show that the macronucleus is nearly as thick dorso-ventrally as the organism and there remain only very thin layers of cytoplasm above and below, too thin to accommodate a food inclusion of any size. This fact aids greatly in the handling of individuals, as the approximate stage of the life cycle can be determined quickly even under very low magnification and the organism for fixation selected.

Under very high magnification the micronuclei can usually be seen. They appear as colorless spheres just anterior to the macronucleus. The number of micronuclei per organism varies between one and four. A careful count in all the organisms examined showed the following frequencies of occurrence: 2 micronuclei — 56 %; 3 micronuclei — 31 %; 1 micronucleus — 10 %; 4 micronuclei — 3 %. The size of the resting micronucleus varies from 3 μ to 6 μ.

Vacuoles

Food vacuoles are usually very numerous. They crowd the endoplasm throughout the body and make the study somewhat difficult. They are filled mainly with algae and plankton but in many cases are crowded with sperm cells of the host.

There is one contractile vacuole located in the posterior region on the left side (Text-Fig. 1. *c. v.*). It fills and contracts slowly and evenly in healthy organisms, emptying its contents through the posterior dorsal surface. No permanent opening in the pellicle is visible. DE MORGAN (1925) says he was unable to observe any canals, but I have regularly seen three or four narrow canals leading into the vacuole. They are irregular in outline and wind in and out among the smaller, non-contractile vacuoles of that region. They are visible only after the discharge of the contractile vacuole and during the early period of its formation.

In moribund organisms the contractile vacuole ceases to function and the small non-contractile vacuoles are seen to fuse together and in turn fuse with the contractile vacuole, forming one huge vacuole which remains until death.

Observations on fixed material.

Neuromotor apparatus

Peripheral system. The pattern of the ciliary rows is depicted in Pl. 1 Figs. 2 and 3. The rows are very numerous and close together. They originate from a transverse fiber in the anterior ventral region (Text-Figs. 1 and 2, *ant. fib.*). The rows of cilia are marked by their basal bodies and longitudinal fibers (Text-Figs. 2 and 3, 1. *cil. fib.*) and pass around the organism in almost parallel rows. Thus the longitudinal ciliary fibers originate at the anterior fiber, pass forward and up over the anterior end, back over the dorsal surface, and down over the posterior end returning on the ventral surface to the anterior fiber. I have never observed any break in these fibers at the posterior end except at division.

Each cilium arises from a basal body situated just under the groove of the pellicular folds. In thin sections "ciliary rootlets" (Rees, 1922) can be seen extending inward from each basal body toward the endoplasm, where they are lost (Pl. 1 Fig. 4). There appear to be no transverse fibrils or commissural fibers, such as were described by Pickard (1927) for *Boveria*. A diagrammatic representation of these parts will be found in Text-Fig. 4.

From the foregoing account it can be seen that the peripheral cilia are all definitely connected to each other by way of the anterior fiber. This fiber, in turn, is directly connected with the peristomal cilia.

Peristomal and oral apparatus. Extending into the endoplasm and following the posterior line of the cytopharynx is found an elongate mass of homogeneous material (Text-Figs. 1 and 2, *mot.*; Pl. 1 Fig. 2). This structure is thicker at its outer end and has a knob on its posterior side. It tapers inward in a slightly curved manner. It stains black with haematoxylin and bright red with Mallory's triple stain. From its position, staining reaction, and connections I believe this to be the motorium and comparable to the motorium described by Sharp (1914), Yocom (1918) and Campbell (1926) and to the neuromotorium of Pickard (1927), Lynch (1929) (1930) and MacClennan and Connell (1931). The motorium measures from 8 μ to 10 μ in length.

The motorium continues into the endoplasm as a relatively heavy strand which seems to be made up of fibers. This strand (Text-Figs. 1 and 2, *ph. st.*) follows the posterior margin of the gullet and frays out in the endoplasm well toward the left side of the organism,

where the individual fibers are lost. This strand is very similar in origin and position to the pharyngeal strand of *Eupoterion pernix* as described by MACCLENNAN and CONNELL (1931). Comparable strands have been described in a number of ciliates (CALKINS and BOWLING, 1929; PICKARD, 1927; etc.).

From the blunt outer end of the motorium two distinct fibers originate. One dips sharply posteriorly and joins the basal elements of the posterior brush while the other is the fiber of the dorsal peristomal row of cilia. This fiber links the basal elements of that row together and proceeds anteriorly to join the anterior fiber (Text-Figs. 1 and 2).

At right angles to the long axis of the motorium, arising from the blunt end and extending in an anterior direction is a solid, deeply staining band of material passing under the large oral brush. This represents the fused basal elements of the oral cilia. From its anterior end it gives off a fiber which joins with the basal elements of the anterior and middle oral brushes. Lining the cytopharynx and originating from the basal element of the large oral brush many fine pharyngeal fibers (Text-Figs. 1 and 2, *ph. fib.*) connect with the motorium along its anterior side.

One more fiber completes the peristomal apparatus. This is the fiber of the ventral peristomal row. It originates at the posterior tip of the anterior fiber, where it is joined by the dorsal peristomal fiber. This ventral peristomal fiber runs under the basal elements of the long ventral peristomal cilia, curves slightly to the outside and ends just anterior to the oral brush.

Structure of peristomal and oral cilia. The nature of the entire set of peristomal and oral cilia can be seen clearly in sectioned material. When thin cross sections are studied a surprising thing is noted. Although the cilia are seen to be single the basal bodies are seen to have fused to form plates. The largest of these plates is that of the oral brush. The plate is of a homogeneous structure staining intense black with haematoxylin and red with MALLORY'S triple stain. Smaller plates of the same nature are found at the bases of the anterior, middle and posterior brushes. The peristomal rows of cilia are likewise seen to be embedded in plates. Pl. 1 Figs. 5, 6 and 7 are of cross sections through the peristomal and oral region of two organisms. Fig. 5 is of a 5μ section through the anterior end of the peristome of an organism fixed in ZENKER'S fluid and stained in MALLORY'S triple stain. Figs. 6 and 7 are of 5μ sections through the lower peristomal and oral region of an

organism fixed in CHAMPY'S fluid and stained with a 10 % solution of acid fuchsin. The motorium and an end of the large oral plate are seen in Fig. 7.

The fusion of the basal bodies into plates is very unusual in a holotrichous ciliate. REES (1922) described a somewhat similar circumstance in *Paramaecium*. He described and figured what he called "paint-brush-like membranelles" in the cytopharynx. The term "membranelles" applied to unfused cilia does not seem acceptable. I should use the less committal term pseudomembranelles for these unique structures.

Discussion of neuromotor apparatus.

In résumé it is seen that the neuromotor apparatus of *Conchophthirius mytili* is a singularly complete and integrated system. Every basal body of the peripheral system is connected through the longitudinal fibers with the anterior fiber. This, in turn, is connected with the peristomal fibers, one of which originates in the motorium. The brushes of cilia about the mouth are seen to connect directly with the motorium but not with the peristomal fibers. This lack of connection is noted in the movements of the cilia, the peripheral and peristomal cilia beating regularly, continuously and metachronously while the beating of the oral brushes is non-continuous and synchronous.

Arguments for a neuroid function of the fibrillar system of ciliates have been given by SHARP (1914), YOCOM (1918), TAYLOR (1920), MACDOUGALL (1928), LYNCH (1930) and MAC CLENNAN and CONNELL (1931). My observations on the neuromotor apparatus of *Conchophthirius mytili* lead me to agree with this conception as opposed to the views of REICHENOW (1927) and others that the fibrillar systems of ciliates are to be considered as either myonemes or skeletal (supportive) structures.

Cytoplasmic inclusions.

The use of Janus green B (HOESCHT) in dilute concentrations (1:50 000) as a vital stain gives a very slow reaction. At first the cytoplasm appears perfectly clear. Gradually, however, inclusions in the forms of rods and incomplete coils are selectively stained a bluish tint. These are the mitochondondria. At best a very incomplete picture of the form and location of the mitochondria can be obtained by the use of vital dyes in this organism. Its value lies in checking the nature of the inclusions, as Janus green B

(HOESCHT) is the most specific of mitochondrial stains, according to COWDRY (1918).

In sections of 5 μ or less after appropriate fixation and staining the mitochondria are easily demonstrated. Best results were obtained with the method of Benda (sulph-alizarinate crystal violet) which I used after fixation for three days in CHAMPY's fluid. The nuclei stained brown, the cytoplasm a reddish tone, while the mitochondria took a vivid purple. In form they are rods or loose coils (Pl. 1 Fig. 8). Many of the rods are oriented toward the periphery of the cell forming a loose layer. The mitochondria of the inner endoplasm show no orientation. Cases of apparent fission of the rods are seen in every section.

The methods of REGAUD, BENSLEY and KULL all give positive results although not as striking as the BENDA method.

The mitochondria lose their capacity to stain after fixatives containing acetic acid.

I found no orientation of the mitochondria about the macronucleus as described by HORNING (1926) (1927) in *Paramaecium* and *Nyctotherus*. Only rod-like mitochondria and loose coils were found in *Conchophthirius mytili* and never spheres, as described by CAUSEY (1925) (1926).

The use of neutral red, as noted above, stained only the trichocysts. The cytoplasm remained perfectly clear. No bodies corresponding to the GOLGI bodies of LYNCH (1930) or of HALL (1930) were demonstrated by this method.

In the cytoplasm just anterior to the macronucleus many minute (0.5 μ) spheres of osmiophyllic material were seen after fixation in CHAMPY's fluid and treatment with a 2 % solution of osmic acid for from five to nine days. These spheres did not bleach out with turpentine. Sectioned material gave no hint as to their finer structure. Whether or not they represent the homologue of the GOLGI bodies described for other protozoa I am unprepared at this time to say.

Binary fission.

I have never found many dividing ciliates in any one mussel, eight to ten being the maximum number and this only occasionally. Because of the ease of observation of the macronucleus in life all except the earliest prophase stages can be selected for fixation with the aid of a dissecting microscope. The various stages are recognizable by the gradual rounding up, elongation and constriction of the macronucleus.

Cytoplasmic structures.

During division the old peristomal region, mouth, cytopharynx and gullet together with the organelles become entirely reorganized. As the macronucleus is elongating the peristomal groove pinches in about half way down its length and the sides fuse. The activity of the peristomal and oral cilia decreases greatly. The old gullet and cytopharynx disappear. The whole peristomal groove seems to become very shallow. Then after the center fusion of the lips of the peristomal groove is complete, the cytoplasm just anterior to this fusion point begins to push in. A similar invagination is taking place in the region of the old mouth. By this time plasmotomy has started with the result that the two developing oral regions are drawn apart. Textfig. 5, *A* shows a sketch of an organism at this stage. Text-Fig. 5, *B* and *C* show sections through the deepest indentation of the mouth regions. The reorganization of the posterior mouth in the general location of the old mouth is a bit in advance of the anterior one. Oral organelles are beginning to form. These organelles are seen to be generalized structures at this stage lacking the differentiation found in the resting organism (compare with Pl. 1 Figs. 5, 6 and 7). The two oral regions and peristomes are completeley reorganized before the daughter chromosomes have separated.

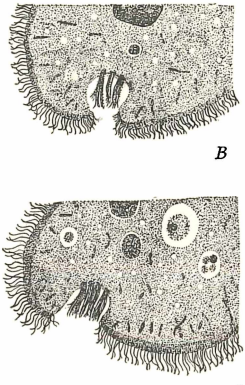


Fig. 5. A., sketch of dividing organism. Lines indicate sections taken. B., a 5 μ cross section through the developing oral region of anterior daughter showing reorganization of organelles in progress. C., a 5 μ cross section through the developing oral region of posterior daughter. The reorganization of the organelles is slightly in advance of that shown in B. B and C drawn with the aid of camera lucida. Sections are viewed from the anterior end of the organism. Zenker's-Heid. haem. $\times$ 1000.

As plasmotomy begins a new contractile vacuole makes its appearance in a position on the left side of the organism opposite the new mouth of the anterior daughter. The contractile vacuole of the posterior daughter is found in the same relative position. Pl. 2 Fig. 9 shows a ventral view of an organism in a late stage of fission. The two new peristomal regions are nearly reorganized and two contractile vacuoles are

present. The line of plasmotomy is clearly marked and the longitudinal ciliary fibers are beginning to break.

Micronucleus.

The first indication of division is to be looked for in the micronuclei. In the resting stage they are seen as more or less compact spheres measuring 3 μ to 6 μ in diameter (Pl. 2 Fig. 10). These spheres start to swell and the chromatin becomes evacuolated (Pl. 2 Fig. 11). A distinct halo appears about the chromatin and the whole micronucleus pulls out somewhat, so that one axis is longer than the other. The chromatin resolves itself into a stranded condition the major part of the strands being oriented parallel to the long axis of the nucleus (Pl. 2 Fig. 12). By this time the micronuclei have attained a size somewhat greater than twice the size of the resting nucleus (12—16 $\mu \times$ 10—11 μ). During this prophase stage the micronuclei migrate from their usual position just anterior to the macronucleus to a position lateral to the macronucleus (Pl. 3 Fig. 19). The micronuclei remain in this spindle shape for a comparatively long period, in fact usually throughout the entire time required to complete the division of the macronucleus. After this "period of waiting" the chromatin threads shorten and condense into sixteen definite though somewhat irregular chromosomes (Pl. 2 Fig. 13). Sections through the equatorial plate show this number quite clearly (Pl. 2 Fig. 14). The chromosomes shorten further and form a definite equatorial plate (Pl. 2 Fig. 15) with irregular spindle fibers clearly visible. The chromosomes always appear to be lined up parallel to the long axis of the spindle. I have never found them shorter than those shown in Pl. 2 Fig. 15. Actual division is probably a rapid process as very few of these stages were found. The ones seen certainly appear as if the division was taking place in a transverse manner (Pl. 2 Fig. 16). This situation parallels that found by TURNER (1930) in *Euplotes patella* and CALKINS (1930b) in *Uroleptus halseyi* but because of the rapidity of the process and the difficulty of observation I can not be absolutely certain of transverse chromosomal division in *Conchophthirius mytili.*

The telophase is again a slow process. The two sets of sixteen chromosomes retain their form long after separation (Pl. 2 Fig. 17) but gradually fuse and contract as the nuclear membrane is pulled out between the two daughter nuclei. This "separation spindle" persists for some time (Pl. 2 Fig. 18) so that by the time it has disappeared the two new micronuclei resemble very closely the resting stage.

The micronuclei included in the posterior daughter organism move over slightly and occupy their normal position anterior to the macronucleus while those micronuclei included in the anterior daughter organism migrate to the anterior cytoplasm.

Throughout the process outlined above the micronuclei, whether two, three or four, are usually found in about the same stage of mitosis. Occasionally, however, slight irregularities occur as seen in Pl. 3 Figs. 22, 23 and 24.

Macronucleus.

Unlike the micronuclei, the macronucleus of *Conchophthirius mytili* during division presents a perplexing and unique problem.

About the time the micronuclei have reached their lateral position the macronucleus rounds up. Then internal differentiation begins to take place. The central portion of the chromatin appears to become more concentrated and becomes surrounded by a lighter band. This band is, in turn, surrounded by a circle of deeply staining chromatin. Toward the periphery of the nucleus another clear area appears. This is shown in Pl. 3 Fig. 19. The nucleus then begins to elongate and the central zone of chromatin seems to contract, the inner clear zone becoming somewhat wider. The outer zone loses its sharp line.

The picture presented at this stage is an elongate nucleus with a compact, densely-staining core of chromatin, surrounded by a lightly staining chromatin area. Around this area and composing the greater part of the nucleus is another region of chromatin which, except for a narrow region near the periphery, takes the stain somewhat less densely than the core. The narrow region or band stains intensely (Pl. 3 Fig. 20).

Subsequent elongation and constriction of the macronucleus result in a gradual fading out of the band and an increased concentration of the central ball (Pl. 3 Figs. 21, 22 and 23). As the two daughter macronuclei separate the central ball of chromatin remains within the nuclear membrane in the division plane. The nuclear membrane pulls out into a long tube which flares at the middle to accommodate the ball of residual chromatin (Pl. 3 Fig. 24). Finally the membrane breaks and the residual ball is left with its covering of the central portion of the tube. This covering shortly disappears and the residual mass starts to disintegrate in the cytoplasm (Pl. 3 Fig. 25). It becomes rough in outline and gets smaller and smaller (Pl. 3 Fig. 26) until, by the end of plasmotomy, no trace of it can be found.

The residual chromatin is absorbed into the cytoplasm of either the anterior or posterior daughter organism.

This chromatin elimination from the macronucleus of *Conchophthirius mytili* is apparently a normal and invariable occurrence. I have always found it in normal vegetative divisions. The amount of chromatin extruded varies but slightly in different individuals. That it is similar to the remaining macronuclear chromatin, at least as far as microchemical test will show, is shown by the FEULGEN nuclear reaction. The whole macronucleus stains a vivid lavender, the concentration of the areas indicated by intensity in color. The residual mass is more intense than any of the rest of the chromatin. This ability to stain after the FEULGEN reaction is maintained until disintegration (or absorption) is complete (see Pl. 3 Fig. 26).

Discussion.

The division of the micronuclei of ciliates seems to follow a rather stereotyped course. The micronuclei of *Conchophthirius mytili* conform to the general type. I was unable, however, to find any trace of centrosomes, as reported by MANWELL (1928) in *Pleurotricha lanceolata*, MAC DOUGALL (1925) in *Chilodon uncinatus* and TURNER (1930) in *Euplotes patella*. What may possibly be an intradesmose is seen regularly in the telophase stages (Pl. 2 Fig. 18). This corresponds, no doubt, to the intradesmose described by WENRICH (1926) in *Paramaecium trichium*.

Very few accounts of chromatin elimination from the dividing macronucleus of ciliates are found in the literature. Aside from the very complete account of this phenomenon given by CALKINS (1930a) for *Uroleptus halseyi* and his briefer account of the same process in *Uroleptus mobilis* (CALKINS, 1919) very little notice has been taken of this type of elimination, if it occurs. ROSSOLIMO and JAKIMOWITSCH (1929) describe and figure elimination of nuclear material from the macronuclei of *Conchophthirius steenstrupii*. This species has seven macronuclei, each of which casts out a small amount of chromatic material at division. DILLER (1928) figures "macronuclear fragments" in the cytoplasm of daughter organisms in his paper dealing with *Trichodina*. He does not refer to these fragments in his discussion. One finds the same situation in the paper by MAC CLENNAN and CONNELL (1931) on *Eupoterion pernix*. This ciliate, belonging to the family Conchophthiridae, has the same type of macronuclear division as *Conchophthirius mytili*, if one is to judge by the authors' figure. In their Fig. 5 is shown a late division stage in which the daughter

macronuclei are nearly separated. The macronuclear membrane is pulled out and surrounds a small mass of material which I assume, from their drawing, to be chromatin. If this is the case then *Eupoterion pernix* exhibits a macronuclear activity parallel to that here described for *Conchophthirius mytili.*

This chromatin elimination from the macronucleus of dividing *Conchophthirius* can be compared only in part to that described in *Uroleptus halseyi* by CALKINS (1930a). I found no substance in the macronucleus that would correspond to the X granules of *Uroleptus.* The chromatin mass stains a brilliant red with the BORREL stain and a deep lavender after the FEULGEN reaction. I have never seen any of the green of the BORREL stain within the macronuclear membrane of vegetative organisms. This chromatin mass may be compared, however, to the D granules of *Uroleptus.* These D granules are the chromatin granules sloughed off prior to division.

As to the physiological significance of this casting out of part of the macronuclear chromatin I can not say. CALKINS (1930a) refers to the similar process in *Uroleptus halseyi* as a "purification process". The fact that it occurs with such regularity in normal division might lead one to suppose that the extrusion mass may represent worn out, waste or accumulated residual chromatin, eliminated preliminary to reorganization after division.

Some evidence is at hand to show that this elimination of chromatin occurs in "old" organisms. By "old" I mean the relative length of time after conjugation. One conjugation epidemic was found ((Fall) 1931, details to be given in the second paper of this series). At the onset of the epidemic of conjugation (October 25, 1931) a few normal divisions were found, all showing chromatin elimination in the usual manner. Many conjugating pairs were found but no exconjugant reorganizations. Subsequent collections (October 26, 27, 29, November 1, 3, 7, 10 and 14) gave few conjugating pairs, many exconjugant reorganizations and a few apparently normal divisions. But in no case did I find chromatin elimination in these dividing forms! Of course I can not be sure that these ciliates had just conjugated. Approximately 90 % of the organisms, however, were exconjugants in various stages of reorganization. From that date until December 31 I found no chromatin elimination in vegetative division. Collections made that day gave a few divisions showing small extrusion bodies and their occurrence steadily increased thereafter.

Summary.

1. *Conchophthirius mytili* was described as a new species by DEMORGAN (1925). It is a member of the family Conchophthiridae (REICHENOW, 1929), suborder Trichostomina, order Holotrichida. It is found in the mantle cavity of the mussel, *Mytilus edulus*.

2. Observations, during the present study, were made from both living and fixed material.

3. One large macronucleus and from one to four micronuclei are present in each organism.

4. A well integrated neuromotor system is described, consisting of a peripheral apparatus and a peristomal and oral apparatus. A motorium, connected directly with both sets of apparatus, is present.

5. Peculiar sac-like trichocysts are present.

6. Mitochondria and osmiophyllic bodies are described.

7. At division the old cytopharynx, gullet and organelles of that region disappear and two new mouths appear.

8. Division is apparently initiated by the macronuclei. These conform to the usual ciliate type and at mitosis sixteen chromosomes are formed on the metaphase plate.

9. The dividing macronucleus is peculiar in that it casts out a part of its chromatin at every vegetative division, with the exception of a few divisions subsequent to reorganization after conjugation. This residual mass of chromatin is broken down and disappears in the cytoplasm of either daughter organism.

Literature cited.

ANDRE, EMILE (1910): Sur quelques Infusoires marins parasites et commensaux. Rev. Suisse Zool. Vol. 18 p. 173—187.

CALKINS, GARY N. (1919): Uroleptus mobilis ENGELM. I. History of the nuclei during division and conjugation. Journ. Exp. Zool. Vol. 27 p. 293—357.

— (1930a): Uroleptus halseyi CALK. II. The origin and fate of the macronuclear chromatin. Arch. f. Protistenk. Bd. 69 p. 151—174.

— (1930b): Uroleptus halseyi CALK. III. The kinetic elements and the micronucleus. Ibid. Bd. 72 p. 49—70.

CALKINS, CARY N. and BOWLING, RACHEL (1928): Studies on Dallasia frontata STOKES. I. Polymorphism. Biol. Bull. Vol. 55 p. 101—112.

CAMPBELL, ARTHUR SHAKELTON (1926): The cytology of Tintinnopsis nucula (FOL) LAACKMANN, with an account of its neuromotor apparatus, division, and a new intranuclear parasite. Univ. Calif. Publ. Zool. Vol. 29 p. 179—236.

Causey, David (1925): Mitochondria and Golgi bodies in Entamoeba gingivalis (Gros) Brumpt. Univ. Calif. Publ. Zool. Vol. 28 p. 1—18.

— (1926): Mitochondria in Noctiluca scintillans (Mac Cartney, 1810). Ibid. Vol. 28 p. 225—230.

Cowdry, E. V. (1918): The mitochondrial constituents of protoplasm. Carnegie Institution of Wash. Publ. Vol. 8 p. 41—160.

De Morgan, W. (1925): Some marine ciliates living in the laboratory tanks at Plymouth. Journ. Marine Biol. Assoc. of United Kingdom Vol. 13 (n. s.) p. 600—658.

Diller, William F. jr. (1928): Binary fission and endomixis in the Trichodina from tadpoles (Protozoa, Ciliata). Journ. Morph. and Physiol. Vol. 46 p. 521—561.

Ehrenberg, C. G. (1838): Die Infusionsthierchen als vollkommene Organismen. Ein Blick in das tiefere organische Leben der Natur p. xviii—547. Leipzig.

Engelmann, W. (1862): Zur Naturgeschichte der Infusionsthiere. Zeitschr. f. wiss. Zool. Bd. 11 p. 347—393.

Hall, R. P. (1930): Cytoplasmic inclusions of Trichamoeba and their reaction to vital dyes and osmic and silver impregnation. Journ. Morph. Vol 49 p. 139—151.

Horning, E. S. (1926): The studies on the mitochondria of Paramecium. Australian Journ. Exp. Biol. and Med. Sci. Vol. 3 p. 89—94.

— (1927): Mitochondrial behavior during the life-cycle of Nyctotherus cordiformis. Ibid. Vol. 4 p. 69—74.

Klein, B. M. (1926): Über eine neue Eigentümlichkeit der Pellicula von Chilodon uncinatus Ehrbg. Zool. Anz. Bd. 67 p. 160—162.

Lynch, James E. (1929): Studies on the ciliates from the intestine of Strongylocentrotus. I. Entorhipidium gen. nov. Univ. Calif. Publ. Zool. Vol. 33 p. 27—56.

— (1930): Studies on the ciliates from the intestine of Strongylocentrotus. II. Lechriopyla mystax gen. nov., sp. nov. Ibid. Vol. 33 p. 307—350.

Mac Dougall, M. S. (1925): Cytological observations on Gymnostomatous ciliates, with a description of the maturation phenomena in diploid and tetraploid forms of Chilodon uncinatus. Quart. Journ. Micr. Sci. Vol. 69 p. 361—384.

— (1928): The neuromotor apparatus of Chlamydodon sp. Biol. Bull. Vol. 54 p. 471—484.

Mac Clennan, R. E. and Connell, F. H. (1931): The morphology of Eupoterion pernix gen. nov., sp. nov. A holotrichous ciliate from the intestine of Acmaea persona Eschscholtz. Univ. Calif. Publ. Zool. Vol. 36 p. 141—156.

Manwell, Reginald D. (1928): Conjugation, division, and encystment in Pleurotricha lanceolata. Biol. Bull. Vol. 54 p. 417—462.

Pickard, Edith A. (1927): The neuromotor apparatus of Boveria teredinidi Nelson, a ciliate from the gills of Teredo navalis. Univ. Calif. Publ. Zool. Vol. 29 p. 405—428.

Rees, Charles W. (1922): The neuromotor apparatus of Paramaecium. Univ. Calif. Publ. Zool. Vol. 20 p. 333—364.

Reichenow, Edward (1927—1929): Doflein's „Lehrbuch der Protozoenkunde" (ed. 5). p. viii—1262. Jena.

Rossolimo, L. L. and Frau Jakimowitsch, K. (1929): Die Kernteilung bei Conchophthirius steenstrupii St. Zool. Anz. Bd. 84 p. 323—333.

SHARP, ROBERT G. (1914): Diplodinium ecaudatum with an account of its neuromotor apparatus. Univ. Calif. Publ. Zool. Vol. 13 p. 44—122.

STEIN, F. (1861): Über ein neues parasitisches Infusor aus dem Darmkanal von Paludina. Sitz.-Ber. d. k. böhm. Ges. d. W. zu Prag (1. Halbj.) p. 85.

TAYLOR, C. V. (1920): Demonstration of the function of the neuromotor apparatus in Euplotes by the method of microdissection. Univ. Calif. Publ. Zool. Vol. 19 p. 403—470.

TURNER, JOHN P. (1930): Division and conjugation in Euplotes patella EHRBG. with special reference to the nuclear phenomena. Univ. Calif. Publ. Zool. Vol. 33 p. 193—258.

WENRICH, DAVID H. (1926): The structure and division of Paramaecium trichium STOKES. Journ. Morph. and Physiol. Vol. 43 p. 81—103.

YOCOM, H. B. (1918): The neuromotor apparatus of Euplotes patella. Univ. Calif. Publ. Zool. Vol. 18 p. 337—396.

Explanation of Plates.

Plate 1—4.

All figures are of *Conchophthirius mytili,* made with the aid of camera lucida, except Fig. 1. Abbreviations for methods of fixation: B. BOUIN's; CH. CHAMPY's; F. FLEMMING's; G.-C. GILSON-CARNOY's; SCH. SCHAUDINN's; S.-A.-Alc. Sublimate-acetic in 95 % alcohol; ZENK. ZENKER's. Abbreviations for the common stains: BOR. BORREL; DEL. DELAFIELD's haematoxylin; FEUL. FEULGEN nuclear reaction; HEID. HEIDENHAIN's haematoxylin; MALL. MALLORY's modified triple stain.

Plate 1.

Fig. 1. Ventral view of vegetative individual drawn from life. The macronucleus appears as a large clear space surrounded by food inclusions. This organism measured 160 $\mu \times 104$ μ.

Fig. 2. Ventral view of organism showing ciliary lines and peristomal apparatus. Cilia have been omitted. F. HEID. $\times 500$.

Fig. 3. Dorsal view of organism. Most of the body cilia have been omitted, their positions indicated by the basal bodies. ZENK. HEID. $\times 500$.

Fig. 4. A 5 μ section through the ectoplasmic region of the ventral surface showing the folds of the pellicle, trichocysts, and basal bodies from which arise the cilia and ciliary rootlets. ZENK. HEID. $\times 2000$.

Fig. 5. A 5 μ cross section through the anterior end of the peristomal groove showing the basal plates of the dorsal and ventral rows of cilia. This section passed through the macronucleus. ZENK. MALL. $\times 1000$.

Fig. 6. A 5 μ cross section through the peristomal groove just anterior to the mouth. A basal plate of the dorsal peristomal ciliary row is shown. Also the basal plate of the middle oral brush. Many food inclusions and mitochondria are shown in the cytoplasm. Ch. Acid fuchsin. $\times 1000$.

Fig. 7. A 5 μ cross section through the motorium. A portion of the large oral brush is included. Also a basal plate of the dorsal peristomal ciliary row. This section is from the same organism as Fig. 6. Ch. Acid fuchsin. $\times 1000$.

Fig. 8. A 5 μ cross section showing mitochondria. Many rods are oriented toward the periphery. A few cases of apparent fission occur. Some of the rods have been cut so that they appear as spheres. Ch. Benda (sulph-alizarinate crystal violet). $\times$ 1500.

Plate 2.

Micronuclei in division.

Fig. 9. Ventral view of a late division stage. The two new oral regions are nearly reorganized. Two contractile vacuoles are present. The macronucleus has divided leaving the ball of residual chromatin. The micronuclei are in late prophase. B. Heid. $\times$ 350.

Fig. 10. Resting micronucleus. Chromatin clumped. B. Heid. $\times$ 2000.

Fig. 11. Early prophase. The chromatin is evacuolated and takes the stain less intensely. Swollen condition. B. Heid. $\times$ 2000.

Fig. 12. Late prophase. The chromatin is in a stranded condition. This represents the stage in which the micronuclei pass through the "waiting period". B. Heid. $\times$ 2000.

Fig. 13. Sixteen long chromosomes have formed. Spindle fibers can be seen. B. Heid. $\times$ 2000.

Fig. 14. Optical section of a micronucleus in the same stage as Fig. 13. The sixteen chromosomes can be seen clearly. S. A. Alc. Feul. $\times$ 2000.

Fig. 15. Metaphase plate. The chromosomes have shortened. B. Heid. $\times$ 2000.

Fig. 16. Early anaphase. Sixteen daughter chromosomes going to each pole. Sch. Bor. $\times$ 2000.

Fig. 17. Late anaphase. Chromosomes becoming rough. Nuclear membrane is being pulled out prior to constriction. B. Heid. $\times$ 2000.

Fig. 18. Telophase. Chromosomes fusing. Long "separation spindle" pulled out between the daughter nuclei. A band of deeply staining material within the separation spindle. This may be an intradesmose. B. Heid. $\times$ 2000.

Plate 3.

Macronuclei in division. All Figs. $\times$ 660.

Fig. 19. Early stage in the formation of the residual ball of chromatin. Micronuclei in prophase. Sch. Feul.

Fig. 20. Macronucleus elongate. Residual mass more compact. The dark band near the periphery is characteristic of this stage. B. Heid.

Fig. 21. Later stage in the concentration of the residual chromatin. G.-C. Heid. Orange G counterstain.

Fig. 22. Macronucleus pulling out and the band of concentrated chromatin near the periphery growing fainter. One micronucleus in metaphase. B. Heid.

Fig. 23. Constriction of macronucleus. Residual mass of chromatin in its characteristic position between the separating daughter macronuclei. Two micronuclei, one precociously divided, the other still in prophase. G.-C. Bor.

Fig. 24. Daughter macronuclei pulled apart each with characteristic projection of chromatin ending at residual mass. Nuclear membrane still intact. Two micronuclei, one in telophase, the other late prophase. G.-C. Del.

Fig. 25. Macronuclear membrane has broken and the residual mass of chromatin is starting to disintegrate, evidenced by its rough outline. The division plane has appeared in the cytoplasm. Both micronuclei in late telophase. S. A. Alc. Heid.

Fig. 26. Plasmotomy nearly complete. Macronuclei still show chromatin projections. Residual chromatin has nearly disintegrated in the cytoplasm. SCH. FEUL.

Plate 4.

Photomicrographs.

Fig. 27. Dorsal view of dividing ciliate. The residual mass well formed in the macronucleus. Two micronuclei at the left of the macronucleus. B. HEID. × 265.

Fig. 28. Constriction of the macronucleus leaving the residual mass in the division plane. G.-C. BOR. × 597.

Fig. 29. Ventral view of dividing ciliate. The residual mass is seen just between the daughter macronuclei. G.-C. DEL. × 265.

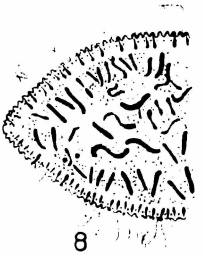
Fig. 30. Same stage as Fig. 29. The strands of chromatin connecting the residual mass with the daughter macronuclei plainly visible S. A. Alc. FEUL. × 265.

Fig. 31. Same stage as Fig. 29. Residual mass between daughter macronuclei. One micronucleus in late telophase. SCH. HEID. × 1040.

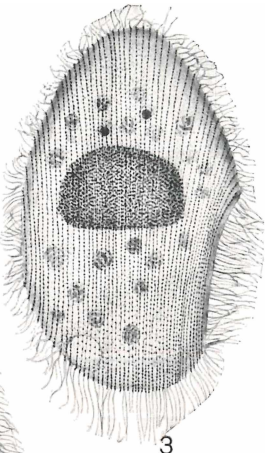
Fig. 32. Same stage as preceeding figure. Residual mass in clear space, surrounded by part of the macronuclear membrane. SCH. HEID. × 1040.

Fig. 33. Later stage in division. Ventral view. The residual mass is disintegrating. The remnant is seen slightly to the right of center. Two daughter micronuclei can be seen near the daughter macronuclei. SCH. FEUL. × 265.

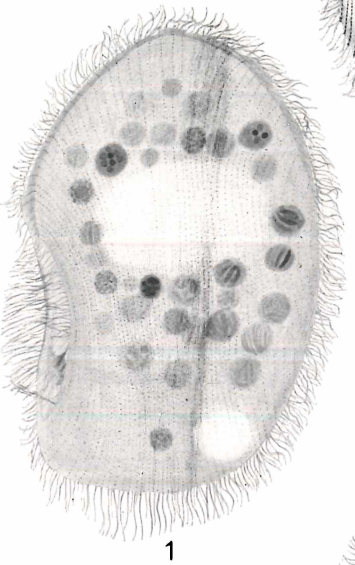
2



8

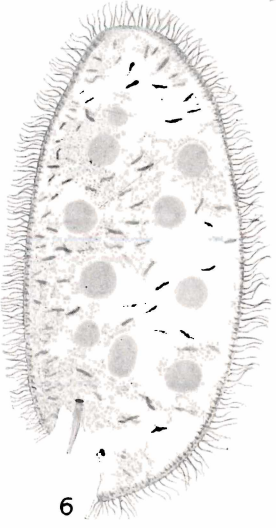


3



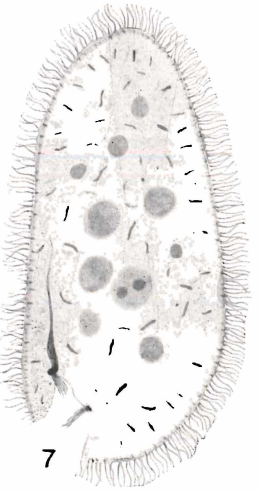
1

4



6

5



7

Kidder I.

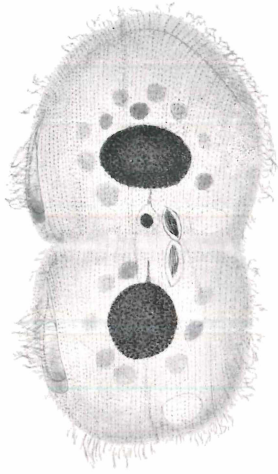
VERLAG VON GUSTAV FISCHER IN JENA.

10

11

12

13



9

14

15

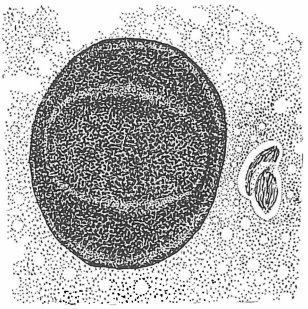
16

17

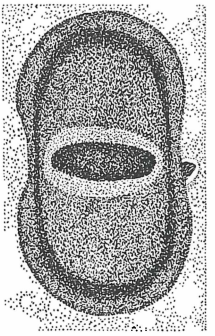
18

Kidder I.

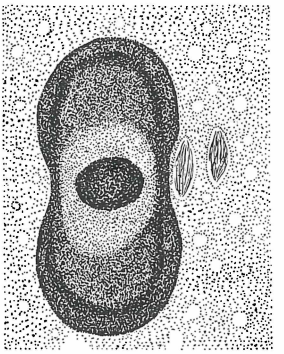
VERLAG VON GUSTAV FISCHER IN JENA



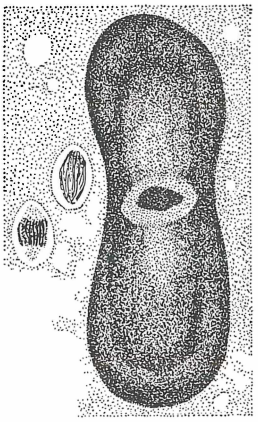
19



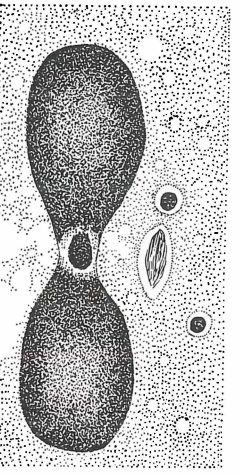
20



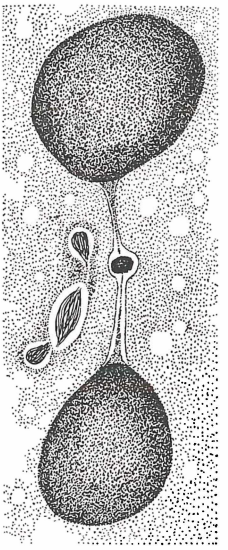
21



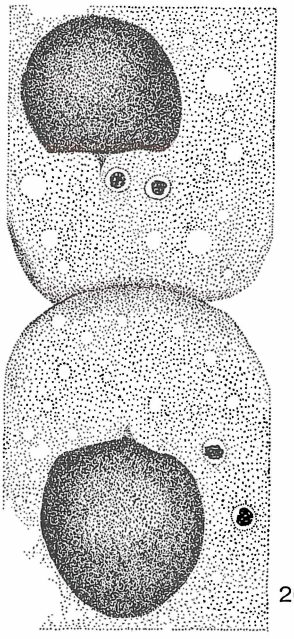
22



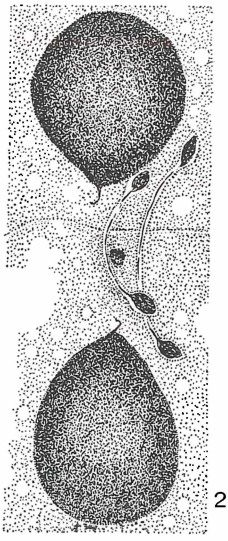
23



24



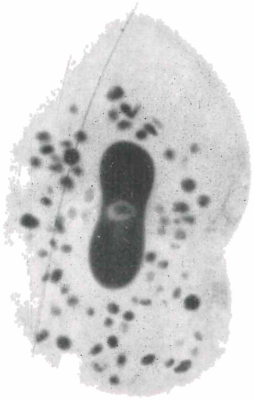
26



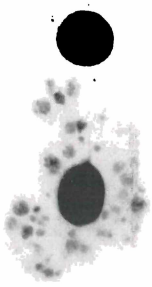
25

Kidder I.

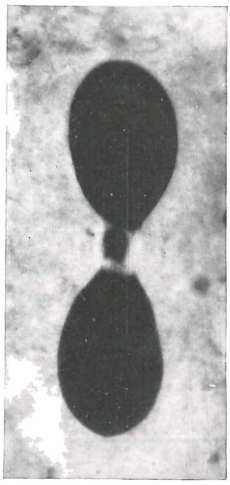
VERLAG VON GUSTAV FISCHER IN JENA



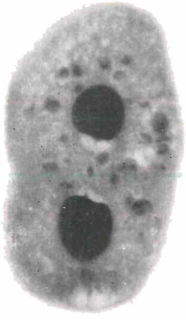
27



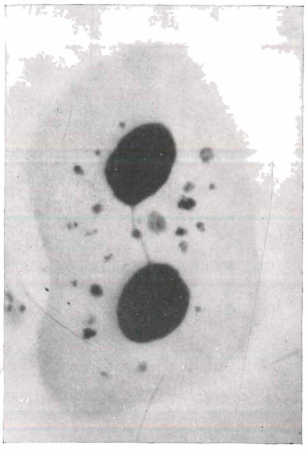
33



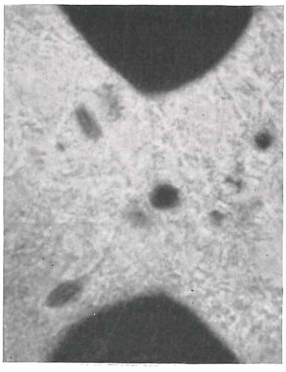
28



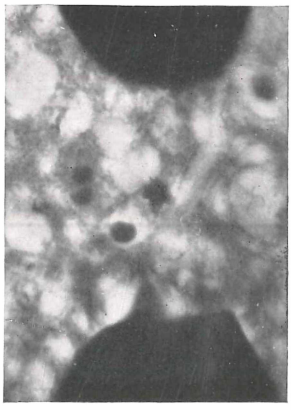
29



30



31



32

Kidder I.

VERLAG VON GUSTAV FISCHER IN JENA