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STUDIES ON CILIATES OF THE FAMILY ANCISTROCOMIDAE CHATTON AND LWOFF (ORDER HOLOTRICHA, SUBORDER THIGMOTRICHA)

I. HYPOCOMINA TEGULARUM SP. NOV. AND CREBRICOMA GEN. NOV.

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#### Introduction

The genus Hypocomina was proposed by Chatton and Lwoff (1924) to include a single species, Hypocoma patellarum Lichtenstein (1921), parasitic on the pallial branchiae of Patella caerulea L. Subsequently, Raabe (1934) described as Hypocomina carinata a ciliate from Mytilus edulis L. which Jarocki (1935) pointed out obviously does not belong to the genus Hypocomina. Raabe used the name Hypocomina carinata again in 1938, however, and no literature has come to my attention in which the form in question is assigned to another genus. In the present paper I will describe as Hypocomina tegularum sp. nov. an ancistrocomid ciliate from the ctenidium of Tegula brunnea (Philippi); and on the basis of my studies on the morphology of a ciliate which I presume to be identical with "Hypocomina" carinata, I will establish the position of Raabe's species in a new genus, Crebricoma.

HYPOCOMINA TEGULARUM SP. NOV.

(Plate I, Figs. 4, 5)

The body is elongated and compressed dorso-ventrally. Fifteen living individuals taken at random ranged in length from 26  $\mu$  to 36  $\mu$ , in width from 12  $\mu$  to 17  $\mu$ , and in thickness from 9  $\mu$  to 12  $\mu$ , averaging about 31  $\mu$  by 15  $\mu$  by 11  $\mu$ . As seen in dorsal view the curvature of the left margin of the ciliate is a little more pronounced than that of the right margin, although this asymmetry is not conspicuous and in most fixed specimens is barely apparent. The body is widest near the middle and is rounded posteriorly. The anterior end is attenuated, bent ventrally, and truncate at the tip. The reduced ciliary system, to be described presently, is disposed in a shallow, relatively flat depression occupying the anterior half of the ventral surface; the dorsal surface and that part of the ventral surface posterior to the thigmotactic field are convex.

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A contractile suctorial tentacle enables the ciliate to attach itself to the epithelial cells of the ctenidium of the host and to suck out their contents. I have not yet succeeded in determining the exact nature of the tentacle in the ancistrocomid ciliates which I have examined and the accounts of other authors regarding its structure are likewise unsatisfactory. In Hypocomina tegularum, it appears to be a membranous tube-like extension of the attenuated anterior end which is protracted and contracted at will. When fully extended the tentacle is about  $3 \mu$  in length and  $1-1.5 \mu$  in diameter. It is hoped that further studies on ancistrocomid ciliates will shed some light on the problem of the structure of this interesting organelle.

The internal tubular canal continuous with the suctorial tentacle is difficult to study in the living ciliates, but in material stained with iron hematoxylin or alum hematoxylin it can usually be traced posteriorly for a distance equal to about one-third the length of the body (Plate I, fig. 4, c). In most individuals it is directed obliquely to the right. In fixed specimens the extreme anterior portion of the canal is approximately  $1-1.5 \mu$  in diameter, while the remainder of it appears to be swollen and to have the form of a poorly defined clear area. Lichtenstein (1921) interpreted the comparable swelling of the canal in Hypocomina patellarum to be a "balle alimentaire." Jarocki (1935) attributed the clear area described by Raabe (1934) behind the tentacle of Hypocomella macomae and "Hypocomina" carinata to hyperdistension of the canal due to unequal infiltration by the fixing reagent or as a result of its action upon moribund, partly plasmolyzed individuals. It is interesting to note in this connection, however, that some ancistrocomid ciliates rarely show this phenomenon upon fixation and the canal can be traced to within a short distance of the posterior end of the body.

The delicate thigmotactic cilia of Hypocomina tegularum are about  $6-7 \mu$  in length and are disposed in nine longitudinal rows. All the rows originate about  $3 \mu$  or  $4 \mu$  posterior to the base of the suctorial tentacle and are about one-half the length of the body (Plate I, fig. 4). The first five rows from the right side of the ciliate are appreciably longer and sometimes appear to lie closer together than the other four rows. Chatton and Lwoff (1924) reported that in Hypocomina patellarum a ridge-like eminence (carène) divides the ciliary area into two unequal fields. These authors did not, however, state how many of the ten ciliary rows described by Lichtenstein (1921) for H. patellarum are in each complex and did not give any facts concerning the relative lengths of the rows comprising the two fields. In Hypocomina tegularum I have not discerned an unmistakable eminence separating the five rows on the right from the four rows on the left, although in some living and fixed individuals the distance between the distal portions of the fifth and sixth rows is somewhat greater than the distance between the other rows.

## EXPLANATION OF PLATE I

FIGURE 1. Crebricoma carinata (Raabe) comb. nov. Ventral aspect. Schaudinn's fixative-iron hematoxylin. Drawn with aid of camera lucida. × 1130.

FIGURE 2. Crebricoma carinata (Raabe) comb. nov. Dorsal aspect, from life.

FIGURE 3. Crebricoma carinata (Raabe) comb. nov. Lateral aspect from right side, from life.

FIGURE 4. Hypocomina tegularum sp. nov. Ventral aspect. Schaudinn's fixative-iron hematoxylin. Drawn with aid of camera lucida.  $\times$  1870.

FIGURE 5. Hypocomina tegularum sp. nov. Lateral aspect from left side, from life.

c = internal tubular canal, cv = contractile vacuole, fv = food vacuole, ma = macronucleus, mi = micronucleus.

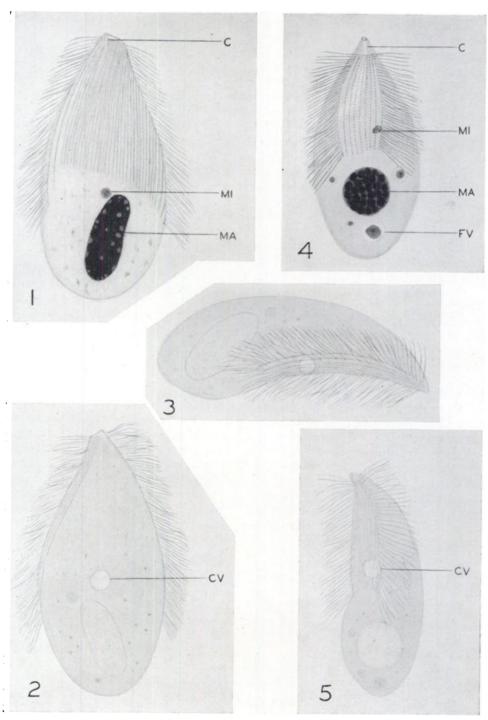


Plate I

The cytoplasm is colorless and contains numerous food inclusions and refractile granules. The refractile granules are apparently lipoid droplets, as they are dissolved out by toluol used for clearing following staining. One or more large food vacuoles (Plate I, fig. 4, fv) are usually present in the posterior part of the body. The contractile vacuole (Plate I, fig. 5, cv) lies near the middle of the body and opens to the exterior on the ciliated ventral surface. I have not detected a permanent opening in the pellicle.

The spherical macronucleus (Plate I, fig. 4, ma) is situated in the posterior half of the body. In preparations stained with iron hematoxylin or alum hematoxylin the chromatin appears to be aggregated into many large, round granules. In ten individuals fixed in Schaudinn's fluid and stained with iron hematoxylin the diameter of the macronucleus ranged from  $5.5 \mu$  to  $7 \mu$ .

The spherical or ovoid micronucleus (Plate I, fig. 4, mi) is usually situated anterior to the middle of the body. It is very difficult to detect in living individuals. In stained preparations the chromatin is dispersed into granules of varying size and shape lying for the most part near the periphery. In ten individuals fixed in Schaudinn's fluid and stained with iron hematoxylin the diameter of the micronucleus ranged from  $1.6 \mu$  to  $2.2 \mu$ .

I found Hypocomina tegularum to be present in small numbers on the ctenidium of two of three specimens of Tegula brunnea (Philippi) which I collected at Carmel Bay, California, in September, 1944.¹ Since that time I have had the opportunity to examine a large number of individuals of Tegula brunnea from localities near Moss Beach and Dillon Beach, California, but found none to be infested by Hypocomina tegularum.

Hypocomina tegularum sp. nov.

Diagnosis: Length  $26 \mu$ - $36 \mu$ , average about  $31 \mu$ ; width  $12 \mu$ - $17 \mu$ , average about  $15 \mu$ ; thickness  $9 \mu$ - $12 \mu$ , average about  $11 \mu$ . The anterior end is attenuated, bent ventrally, and provided with a contractile suctorial tentacle continuous with an internal tubular canal. The ciliary rows are nine in number and are disposed in a shallow depression occupying the anterior half of the ventral surface. The first five rows from the right are slightly longer than the other four rows. The macronucleus is spherical and is situated in the posterior half of the body. The micronucleus lies anterior to the middle of the body. Ectoparasitic on the ctenidium of *Tegula brunnea* (Philippi) (Carmel Bay, California). Syntypes are in the collection of the author.

Crebricoma gen. nov.
Crebricoma carinata (Raabe) comb. nov.

(Figure 1. Plate 1, Figs. 1-3)

I have distinguished two species of ancistrocomid ciliates parasitic on the epithelium of the palps and ctenidial filaments of *Mytilus edulis* L. from various localities in San Francisco Bay. One of these is *Hypocomides mytili* Chatton and Lwoff; the other I presume to be identical with "*Hypocomina*" carinata Raabe, for which I propose a new genus, *Crebricoma*. My observations on the morphology

<sup>1</sup> I am indebted to Dr. D. T. MacDougal for his kindness in arranging my trip to Carmel which made possible the collection of the original material of the ciliate described herein.

of this species permit me to augment the original description given by Raabe. It seems advisable, therefore, to present herein a revised description of *Crebricoma carinata*.

The body is elongated and somewhat compressed dorso-ventrally. Fifteen living individuals taken at random ranged in length from  $58 \mu$  to  $71 \mu$ , in width from  $27 \mu$  to  $39 \mu$ , and in thickness from  $22 \mu$  to  $31 \mu$ , averaging about  $64 \mu$  by  $31 \mu$  by  $25 \mu$ . The anterior end is narrowed, bent ventrally, and in dorsal view is seen to end in an oblique truncation, the right side of which projects a little more than the left side (Plate I, figs. 1, 2). The body is widest near the middle and is rounded posteriorly. The ciliary system, to be described presently, is disposed for the most part on the shallow concavity occupying the anterior two-thirds of the ventral surface; the dorsal surface and that part of the ventral surface posterior to the thigmotactic area are convex.

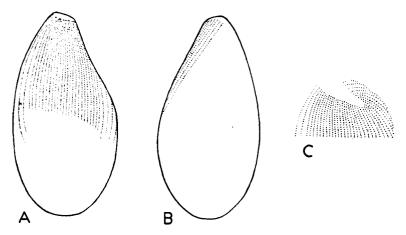


Figure 1. Crebricoma carinata (Raabe) comb. nov. Distribution of ciliary rows. Semidiagrammatic representations based on camera lucida drawings of specimens fixed in Schaudinn's fluid and impregnated with activated silver albumose (protargol). A. Ventral aspect, B. Dorsal aspect, C. View of anterior end.

The suctorial tentacle is situated on the right side of the anterior truncation. I am at present unable to make any conclusive statement with regard to the structure and contractile properties of the tentacle in this species. The internal tubular canal continuous with the tentacle cannot be satisfactorily distinguished in living material, but can be demonstrated in some fixed specimens which have been stained with iron hematoxylin. It appears to pass at first dorsally and then ventrally and obliquely to the right. I have not succeeded in tracing it posteriorly for more than a short distance. A broad lighter area behind the anterior part of the canal is frequently observed in fixed individuals.

As described by Raabe, the ciliary system of *Crebricoma carinata* consists of about twenty closely-set rows bordered on the right by two longer and more widely-spaced rows and on the left by three such rows. According to my observations, however, the more widely-spaced rows on the left side do not represent a complex separate from the closely-set rows. The number of rows in the thigmotactic field

of *C. carinata* is very difficult to determine, since some of the rows on the left are lateral in position and several of them originate on the dorsal surface. In one individual impregnated with activated silver albumose I counted thirty-four rows and in another individual, thirty-six rows. In all specimens which I have examined carefully the number of ciliary rows exceeded thirty-two.

The two widely-spaced rows on the right side of the thigmotactic field originate on the ventral surface near the base of the suctorial tentacle. The outer row is the longer and is about two-thirds the length of the body (Fig. 1A). All the rows of the second complex, with the exception of three or four rows on the extreme left which are about as widely-spaced as the two long rows on the right side, are very closely-set and are one-half to two-thirds the length of the body, becoming progressively longer toward the left. Several of the rows on the left side of the thigmotactic field originate on the left lateral margin or on the dorsal surface (Fig. 1B; Plate I, fig. 2). The arrangement of the ciliary rows at the anterior end thus forms an incomplete suture (Fig. 1C) reminiscent of the anterior field of other holotrichous ciliates.

The cilia of *Crebricoma carinata* are about  $10-11\,\mu$  in length. When the organism is attached to the epithelium of the palps or ctenidial filaments of the host the cilia usually exhibit only a sluggish movement. Occasionally one or two small groups of cilia near the anterior end beat energetically. When dissociated from the host *C. carinata* swims actively and its cilia beat metachronously.

Raabe proposed the specific name carinata in allusion to a keel-like prominence on the dorsal surface. I have seen such prominences on seriously shrunken fixed specimens of Crebricoma carinata and on some living individuals which were obviously undergoing plasmolysis. I have not, however, been able to distinguish them on ciliates freshly removed from the host. Raabe also described a sunken space between the keel-like prominence and the outermost ciliary row on the right side, which he said "vermutlich dem von Chatton und Lwoff bei anderen Hypocomiden beschriebenen 'vestige de frange adorale' entspricht." I do not believe such a depression exists in normal individuals, although I have observed groove-like and slit-like depressions form on the lateral and dorsal surfaces of moribund specimens of Crebricoma carinata, as well as Hypocomides mytili.

The cytoplasm is colorless and contains a few small refractile granules, which presumably are lipoid droplets, and food inclusions. I have not discerned any large food vacuoles in this species. The contractile vacuole (Plate I, fig. 2, cv) opens to the exterior on the ventral surface near the middle of the body. There appears to be no permanent opening in the pellicle.

The sausage-shaped or ovoid macronucleus (Plate I, fig. 1, ma) is situated in the posterior half of the body. In preparations stained with iron hematoxylin or the Feulgen nuclear reaction the chromatin is aggregated into a dense reticulum enclosing vacuole-like clear spaces of varying size. Viewed dorsally, the longitudinal axis of the macronucleus is ordinarily placed obliquely to the longitudinal axis of the body. In most specimens the anterior end of the macronucleus is directed dorsally, while the posterior end is directed ventrally. In ten individuals fixed in Schaudinn's fluid and stained by the Feulgen nuclear reaction the macronucleus ranged in length from  $13 \mu$  to  $24.3 \mu$  and in width from  $5.6 \mu$  to  $11.7 \mu$ .

The spherical micronucleus (Plate I, fig. 1, mi) is readily detected in the living ciliate. It is commonly situated near the dorsal surface close to the anterior end of

the macronucleus. Following fixation the micronucleus stains lightly with iron hematoxylin and the Feulgen reaction. The chromatin appears to be homogeneous. In ten individuals fixed in Schaudinn's fluid and stained by the Feulgen reaction the diameter of the micronucleus ranged from  $2.7 \mu$  to  $3.6 \mu$ .

Crebricoma gen. nov.

Diagnosis: The body is elongated, somewhat flattened dorso-ventrally, and narrowed anteriorly. The anterior end is provided with a contractile suctorial tentacle continuous with an internal tubular canal. The ciliary rows are numerous and except for some which originate on the dorsal surface or left lateral margin are disposed on the ventral surface. Two long, widely-spaced rows on the right side of the ciliary system appear to form a complex separate from the remaining rows, which are for the most part closely-set. The arrangement of the ciliary rows at the anterior end of the body forms an incomplete suture. Genotype: Crebricoma carinata (Raabe) comb. nov. (= Hypocomina carinata Raabe).

Crebricoma carinata (Raabe) comb. nov.

Diagnosis: Length  $58 \mu$ - $71 \mu$ , average about  $64 \mu$ ; width  $27 \mu$ - $39 \mu$ , average about  $31 \mu$ ; thickness  $22 \mu$ - $31 \mu$ , average about  $25 \mu$ . Two widely-spaced rows of cilia on the right side, the outer of which is about two-thirds the length of the body, and a series of more than thirty rows, which with the exception of three or four on the left are closely-set, comprise the ciliary system. The closely-set rows are one-half to two-thirds the length of the body, becoming progressively longer toward the left. The macronucleus is sausage-shaped or ovoid. The micronucleus is spherical. Ectoparasitic on the palps and branchial filaments of *Mytilus edulis* L.

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