

Sessiline Peritrichs from the Surface of Some Freshwater Fishes

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Abstract. The paper deals with sessiline peritrichs from the surface of some freshwater fishes. A technique for their observation is described, an emphasis being placed on observation of living ciliates. The importance of individual diagnostic features, among which the pellicular surface structures are also very relevant, is discussed.

The genus *Glossatella* Bütschli is to be rejected as a junior synonym in favour of *Apiosoma* Blanchard. Four species of this genus were observed: a redescription is given of *Apiosoma piscicola* Blanchard and of *Apiosoma campanulata* (Timofeev), together with description of two new species, *Apiosoma phoxini* sp.n. and *Apiosoma gobionis* sp.n.

Epistylis lwoffii F. - F. occurs on fresh-water fishes in four distinct ecoforms on different host species; these are described in detail. They differ chiefly in the form of their stalk.

The relation of sessiline peritrichs to their hosts is discussed. Although they feed on water dispersed particles, their occurrence being influenced by the properties of the fish surface. A marked seasonal variation could be observed, too. Mutual relations of *Apiosoma* and *Epistylis* vary according to the host; either of them may serve as a support for the other one.

A survey of sessiline peritrichs is added, comprising all species recorded hitherto from fishes, together with some critical comments concerning their taxonomic position.

At the present studies of fish parasites proceeding from the purely descriptive, taxonomic stage to the ecology and the host-parasite relations, reveal not only interesting theoretical data, but supply also fundamental knowledge for their more effective control. However, there still remain groups of fish parasites where even the alpha taxonomy is a matter of future studies and where almost no reliable data are available on the occurrence of their species. The sessiline peritrichs from the body surface of marine and freshwater fishes are one of them. Although their presence on the fish is quite common, we find in most of the lists surveying the fish parasitofauna and worked out by different authors, rarely more than the entry "Epizoa spp"; this may cause a serious gap in the conclusions drawn from ecological studies. From the viewpoint of the host—parasite relations these ciliates yield an interesting example of adaptation to parasitic life: in their food requirements they are not dependent on their hosts, feeding on bacteria and particles

37 dispersed in the water, but their association with a given host is determined—in a hitherto enigmatic way—by a complex of properties (mainly chemical) of the host's surface. The result of the interactions of both these environments, of the first and second order, indicate either the abundant or rare presence of these ciliates on the host.

The reason for neglecting the sessiline peritrichs may be due to technical difficulties in their study. In the first place they have to be examined alive since the fixation changes entirely the shape of their body, especially the peristomial area. They are also very sensitive to changed conditions and when removed from the host, soon contract the peristome and perish after a short time. Furthermore, some staining methods, unfamiliar to fish parasitologists have to be employed.

Up to the present, some 35 species of sessiline peritrichs (—not including several synonyms or unsatisfactory descriptions) have been recorded from fishes, the majority of which has been recently described by Soviet authors. (TIMOFEEV 1962, ZHUKOV 1962). With an increased interest in this group, more information is to be expected. The present paper represents an attempt to draw attention to these interesting organisms.

MATERIAL AND METHODS

The sessiline peritrichs were examined in the mucous material scraped from the surface of the fish with a surgical knife. The fresh material was immediately microphotographed because the ciliates die very quickly after the removal from the host. The time of exposure 0.5 to 1 second is sufficient even when using low-speed films of fine grain (Agfa Document) with objective lens 45x, eyepiece 15x. The photos are made with a single lens camera 24x36 mm attached directly to the tubus of the microscope, equipped with a built-in illumination (a 6 W bulb) and making photographing possible under almost field conditions.

The fish should be kept alive during the investigation of the ciliates, because as mentioned before, the ciliates perish very shortly after the fish had been killed and taken out of the water. It is advisable to immobilize the fish by an electric shock (direct net current of 220 V) because otherwise a too firm grasp of the fish may damage the mucous material with the ciliates.

Dry smears are impregnated after Klein's silver method to reveal the argentophilic annuli on the surface, or stained by the Robinow method to reveal the nuclei (in stained *Apiosoma* their shape and dimensions change; the true shape is observable in living specimens only—this can be very easily carried out on dying specimens, where the nucleus becomes visible). Protargol stain was employed to reveal the buccal infraciliature; the method and scheme of designation of its components is the same as used in our previous paper (LOM 1964).

GENUS *APIOSOMA* BLANCHARD, 1883

syn. *Glossatella* Bütschli, 1889

comprises ciliates, living as symphorionts on the surface of water animals, to which they are attached directly by the scopula. The border of this holdfast organelle is equipped with kinetosomes bearing short scopular ciliary organelles. The latter may be absent in adult ciliates. The body has a cylindrical up to elongated

conical shape, the adoral ciliature complying with the general peritrichean scheme. Both the haplo- and polykinety make slightly more than one turn before they plunge into the vestibulum; the three peniculi make a $\frac{3}{4}$ spiral turn downwards the infundibulum, and are situated closely side by side. A compact, triangular macronucleus occupies the position in the aboral half of the body. Only the anterior part of the body is contractile, being capable of drawing the peristomial apparatus inside the body.

The situation within this genus reflects all difficulties of the peritrichean taxonomy, the main problem being, also, uniformity of the body features, the great variability and lack of strict host-specificity.

The body shape: populations which are grouped into different species on the following pages, possess, in most instances, a characteristic body shape. However, it varies largely according to the micro- and macroenvironmental conditions. In weak infections, if we don't find enough ciliates on the fish, we can rarely exactly identify the ciliate. On observing the *Apiosoma*, we must also bear in mind that the ciliate grows and changes its shape during its life cycle. (Fig. 1.) As a rule, the grown-up, or "adult" ciliates reach a very elongated shape; in some populations, however, they remain rather stubby, for unknown reasons. Sometimes their shape may also be altered by the removal from the host. Fixation changes the shape of the ciliate, because the contraction diminishes the body dimensions and also the size and the shape of the macronucleus, which becomes rounded instead of retaining the normal triangular form—the true shape of the body can be ascertained only in living specimens.

The size of the body is very variable, evidently depending on environmental conditions.

The shape of the swarmer is uniformly loaf-like in all species observed.

The position of the micronucleus in relation to the macronucleus is constant enough to serve as a differentiating feature, but the number of possibilities is limited to about four positions: above the macronucleus, beside its upper or bottom part, and below the macronucleus.

The number of argentophilic annuli can be used as species characteristics, taking into account, of course, its variability.

The scopula is—with rare exceptions—of the same structure in all species. Hyaline formations, attached to it and considered as pseudopods in *Apiosoma amoeba* Grenfell, are either mucous secretions of the fish (SHULMAN 1963) or more probably remnants of epithelial cells to which the scopula is firmly attached. The curved borders of the scopula are capable of "biting" into the surface of epithelial cells and often take them along if the ciliate is removed from the skin.

The individual features of *Apiosoma* almost always overlap in different species: therefore a complex of all features of the ciliates has to be estimated before determining it. This should be done with most organisms but here it is of special importance. The use of statistics and large scale investigations will become necessary when more species or different populations will be known.

39 Finally, we have to mention the problem of nomenclature. The genus *Apiosoma* was created by BLANCHARD in 1885 for a ciliate from the skin of a carp, *A. piscicola*. This description is sufficient enough to permit even to-day an identification with ciliates occurring abundantly on carps and other fishes. The genus *Glossatella* was erected by BÜTSCHLI in 1889 for a similar ciliate, described by KENT (1882) under the name *Spirochona tintinnabulum* from the skin of newts. Bütschli was

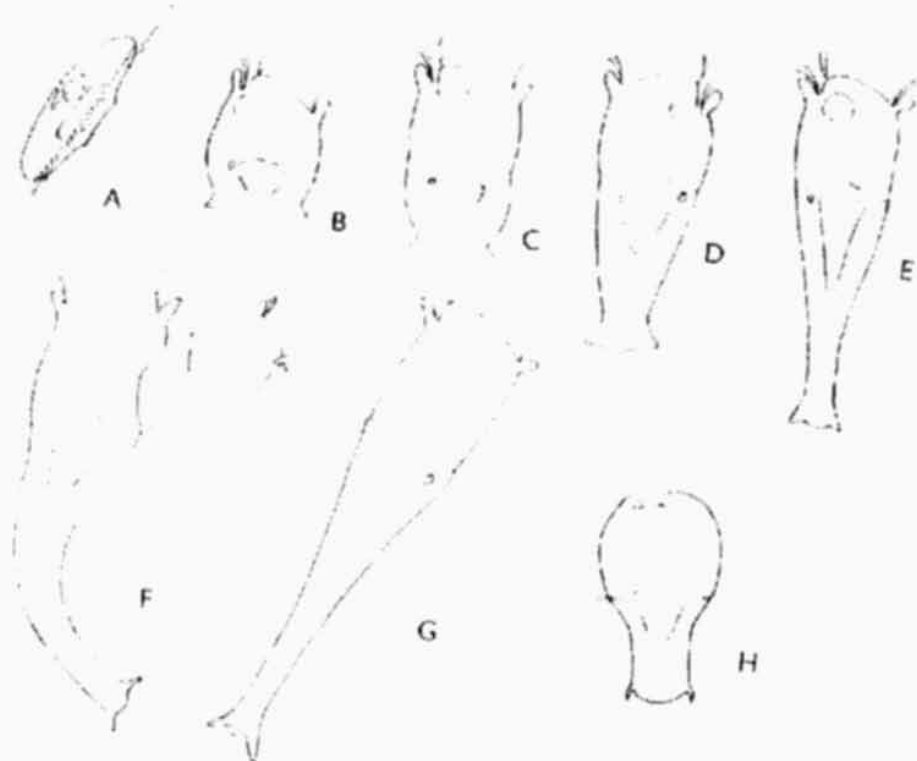


Fig. 1. Morphological changes of *Apiosoma piscicola* during the developmental cycle. The ciliates are drawn schematically in outlines. A—swarmer, B—recently attached ciliate, C+D—the ciliate assumes the "adult", most common shape E, and G. Fig. F shows the size relation of conjugants. H—telotroch formation; under the influence of unfavourable conditions, ciliates of different evolutionary stages are capable to form the telotroch, so that this picture must not be regarded as quite typical.

right in separating *Spirochona tintinnabulum* from the genus *Spirochona*; however, he did not realise, that *Spirochona tintinnabulum* and *Apiosoma piscicola* are congeneric. Therefore, instead of adopting the generic name *Apiosoma* even for *Glossatella tintinnabulum*, he created a new name, *Glossatella*. To that date, three species of the present genus *Apiosoma* were recorded: *Spirochona* (= *Apiosoma*) *tintinnabulum* Kent, 1882, *Apiosoma piscicola* Blanchard, 1885 and *Scyphidia* (= *Apiosoma*) *amoeba* Grenfell, 1887, the latter being unknown to Bütschli. He considered *Apiosoma piscicola* to be most probably a species of *Rhabdostyla*, although he admitted a relation to *Glossatella* because of the pronounced rings on the pellicle. Afterwards, under his influence, the term *Glossatella* was widely accepted; KAHL (1935) coined a new definition for it, while *Apiosoma* is quoted only in an insignificant number of papers. CORLISS (1961) quite correctly quoted *Glossatella* as a probable synonym of *Apiosoma*. We must insist upon the recognition

of the generic name *Apiosoma* as the valid one, the genus *Glossatella* being merely a junior synonym. 40

Apiosoma piscicola Blanchard, 1885

Syn. *Glossatella piscicola* (Blanchard, 1885) Kahl, 1935, *Glossatella cyprini* Šrámek – Hušek, 1953
Found regularly on the body surface of *Cyprinus carpio*, *Abramis brama*, quite often on *Carassius carassius*, *Rhodeus sericeus*, *Leucaspis delineatus*, *Salmo trutta m. fario*, *Gobio Gobio*, *Leuciscus cephalus*, less frequently on *Rutilus rutilus* and *Perca fluviatilis*. TIMOFEEV (1962) found it also on *Barbus barbus* and *Alburnus alburnus* in the USSR.

This species is characterized by the common occurrence of the following features:

1. The body size reaches higher limits than in other species. Populations of the biggest ciliates are found on carps (as big as 40 by 110 μ), populations on other fishes are smaller, the average is e.g., in *Abramis brama* 33 by 80 μ , in *Leucaspis*

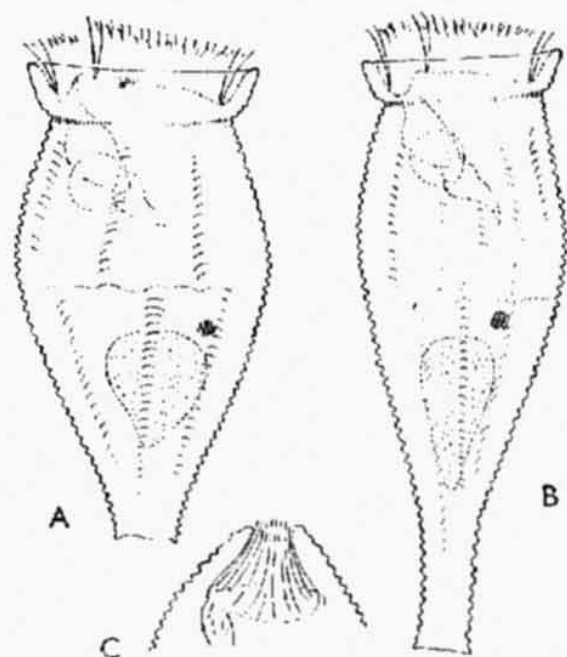


Fig. 2. Body form of *Apiosoma campanulata*. Note the coarse transverse striation of the pellicle and its longitudinal folds. C - contracted peristome retains a conical space under the peristomial lips.

delineatus 38 by 68 μ , in *Gobio gobio* 62 by 25 μ . (The width of the body is measured in the anterior, oral third of the body, the length from the summit of the epistomial disc to the scopula.)

2. Number of the argentophillic annuli on the pellicle tends to be also higher than in other species. The average values (first number indicates the number of annuli between the pectinellar polykinety and the peristome, the second one between the pectinellar polykinety and the scopula) are highest (curiously) in perch 34—52; in trouts they are 37—47; in *Abramis brama* 26—55; in carps 27—49; in *Leucaspis delineatus* 27—49; in *Gobio gobio* 25—50. However, the number of annuli varies considerably, e.g., in one population from *Abramis brama* from 24—27 up to 48—62. The annuli are spaced at intervals slightly superior to one micron.

11 3. Position of the macro- and micronucleus. In grown-up individuals the macronucleus is triangular, its dimensions varying from 15 by 10–11 μ . The rounded till elongated interphasic micronucleus has the medium length of 4 μ and lies closely to the upper part of the macronucleic side (Fig. 1).

4. Shape of the body of grown-up individuals is characteristic—ciliates of other species are not so slender in form as *A. piscicola*. The aboral slender part is usually not thicker than 5 μ .

The buccal ciliary apparatus is shown on Fig. 3A. Since the relatively short

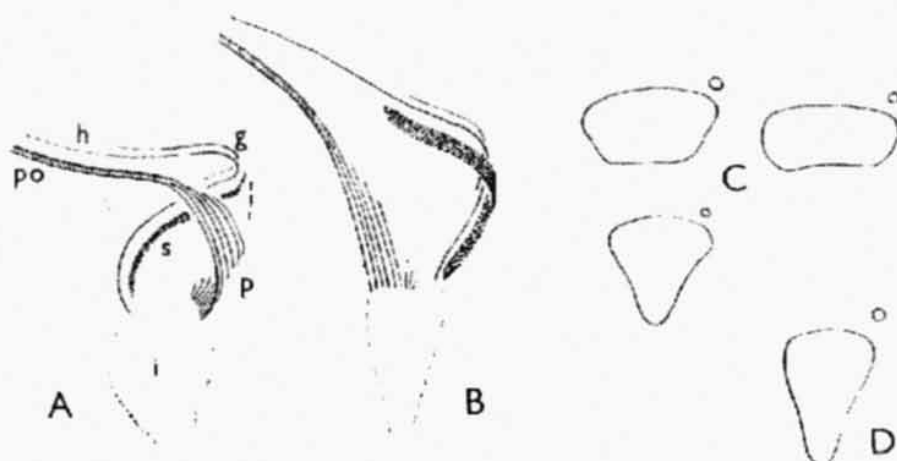


Fig. 3. A—Buccal infraciliature of *Apiosoma piscicola*. The outer spiral of the peristome (adoral zone) is omitted. Terminology used is the same as employed in the previous paper (LOM 1964). B—the same in *A. campanulata*. Note the almost unspiraled course of penicular rows through the infundibulum. h—haplokinety, po—polykinety, g—germinal row, s—impregnable structure, i—infundibulum, p—peniculus-like structure of nine kineties. C—Most frequent nuclear types in *Apiosoma phoxini*. D—Nuclear type of grown up *A. gobionis*.

infundibulum is usually inclined in different angles in relation to the axis of the body; the mutual position of penicular rows and the haplokinety within the infundibulum also varies to a small extent. Sometimes the penicular rows make their turn on the infundibular walls at almost the same level. The epistomial disc is rather flat, only slightly elevated at the side of the infundibulum. Within the peristomial lip there extends an impregnable myofibrillar sphincter, the branch of which extends along the beginning of the adoral zone, similarly to *Epistylis lwoffii*, which will be described later. The well marked peristomial lips are 6–7 μ high. The contractile vacuole discharges into the infundibulum. The whole or one half of the body is filled by nutritive vacuoles.

The shape of the body changes during the developmental cycle; many ciliates, however, do not reach the final slender form. Also the scopula changes its shape. Its borders bear short scopular ciliary organelles. The rest of the scopula is free from these elements, with the exception of perhaps a small Ag-impregnable ring in the center of the scopula. The concave scopula of grown-up ciliates has always a projecting, convex center.

We have never been able to observe any cyst formation. Conjugation (mutual size relation of both conjugants see Fig. 1F) is a frequent phenomenon.

Taxonomic affinities. BLANCHARD'S (1885) description — perfect at its time — of *Apiosoma piscicola* permits to identify our species with this one. The general shape of the body, its dimensions and the host are in full agreement.

The species which FAURÉ—FREMIET (1943a) found to be associated with *Erastophrya chattoni* on the skin of *Gasterosteus aculeatus* is certainly not a *A. piscicola*; his brief description does not permit a precise determination; should it be *A. amoeba*? The same refers to *Apiosoma* associated with *Epistylis hvoffi* and described by the same author (1943b).

Glossatella cyprini Šrámek—Hušek, 1953—as far as the unsatisfactory description permits any identification—is to be considered as a synonym of *A. piscicola*.

TIMOFFEV'S (1962) findings agree in general with BLANCHARD'S and our description.

Apiosoma campanulata (Timofeev, 1962) emend.

Commonly found on the skin of *Perca fluviatilis*; seldom on *Rutilus rutilus*, living with the infected perches on the same localities. TIMOFFEV found it also on *Lucioperca lucioperca*.

The principle characteristic feature of this species is its stubby spindle-shaped form. Elongated forms which are not so slender as in the foregoing species, are rather rare. Furthermore, there is a conspicuous circular striation of the pellicle and, in addition, the pellicle is wrinkled in longitudinal furrows; both these features differentiate the species from the others.

The number of circular annuli is 16—23 on the oral, and 17—27 on the aboral half of the ciliate. They are more loosely spaced than in the preceding species; the argentophile pores of the muciferous pellicular tubuli are less frequent than in *Glossatella piscicola* especially in the aboral end.

The shape of the body is evident from Fig. 1A, B. The sequence of developmental stages is similar to *A. piscicola*, and also no cysts were observed. The peristomial lips are slender, but may reach the height of 10 μ . The epistomial disc is almost flat. In animals with a contracted peristome, there is a free conical space above the inverted epistomial disc, covered by the contracted peristomial lips. Such a space has not been observed in contracted individuals of *A. piscicola*. The impregnable myofibril in the peristomial lip is simply circular, without any branch. Dimensions of the living ciliate again vary according to individual populations, their average measures are 30 by 60 μ ; the elongated forms only rarely reach 95 μ . The buccal ciliary apparatus (Fig. 3B) is of the same type as in *A. piscicola*, but the penicular rows within the infundibulum extend mostly inwards in a straight, unspiralled course.

The spherical to oval micronucleus (2.5 by 4 μ) is situated above the triangular macronucleus, shifted to its side. Under the influence of fixation, the macronucleus assumes an irregularly rounded shape. The anterior border of the macronucleus does not even reach the level of the pectinellar kinety.

Taxonomic affinities. Of the species described hitherto, which have a similar

- 43 shape of the body, we shall consider those with the micronucleus in a similar position: *A. minuta* Chen, *A. peculiariformis* Zhukov, *A. campanulata* Timof., *A. conica* Timof. The dimensions of the first one are smaller, the second one is insufficiently described. Of these last two species we consider *A. campanulata* to be identical with our species—rather because of the same host than because of more points of agreement in the original description of *A. campanulata*.

***Apiosoma phoxini* n.sp.**

Occurs on the skin and gills of minnows (*Phoxinus laevis*) from different localities in Bohemia (= terra typica).

This species is characterized principally ad a) by the shape of the body, the posterior, aboral half of which is never so slender as in *A. piscicola* or *A. campanulata* or *A. gobionis* (for the most typical ciliates see Plate III) and ad b) by the pellicle which protrudes in a sort of ledge in the place where the pectinellar fringe is inserted.

Other characteristic features are shown on the microphotographs. The epistomial disc is more or less vaulted, its upper surface surpassing the peristomial lip; it is almost not elevated at the side of the infundibulum. The average body dimensions are 58 by 26 μ in living ciliates.

In ciliates with a contracted peristome, the great conical space limited by the peristomial lips and epistomial disc is much more distinct than in *A. campanulata*. The buccal structure is of the same type as in *A. piscicola* with only a small difference, i.e., that these penicular rows are separated, and that the argentophilic impregnable structure lining the infundibular part of the haplokinety is broader. In stout individuals, the macronucleus is rounded, in elongated ones it is triangular, about 17 by 12 μ in size. A spherical to oval micronucleus (diameter 1.5 to 3 μ) is situated at the side of the oral face of the macronucleus.

There are 17 to 23 circular annuli on the oral and 25 to 32 on the aboral half of the body.

Taxonomic affinities. The only recorded *Apiosoma* resembling this species is *A. conica*; however, this species is not characterised precisely enough to be comparable with another species. Moreover, it has another host. Though we know that there is no strict host specificity, from the mentioned reasons we prefer to propose the name *Apiosoma phoxini* n.sp. for this ciliate.—Type slide of this ciliate, as well as type slide containing ciliates of the following new species, *A. gobionis* n.sp., are deposited in the collection of the author.

***Apiosoma gobionis* n.sp.**

Up to date this species has been found only on the body surface of *Gobio gobio* from different localities in Bohemia (= terra typica).

It differs from the previous species mainly in the shape of the body. Most typical are the cylindrical stubby individuals (Pl. II, Fig. 4, 5) with a maximum body width across the peristomial lips which are strongly developed. Their height comes up to 9 μ . The average body dimensions are 48 by 28 μ . The ciliates of an elongated shape resemble those of *A. campanulata*, but are lacking the conspicuous body striation and the longitudinal grooves. Their dimensions reach 23 by 85 μ . The developmental cycle is the same as in foregoing species.

The epistomial disc is only slightly vaulted and slightly elevated on the side of the infundibulum: on the side view it is hidden by peristomial lips even in uncontracted ciliates (Pl. II, Fig. 5). There are 28 to 34 annuli on the oral half, and 27 to 37 on the aboral half of the body. The buccal ciliary apparatus is similar to *A. piscicola*, but the impregnable structure lining the infundibular part of the haplokinety is broader, and the kinetosomes of the G-row are less regularly arranged. The circumperistomial impregnable myofibril is well marked.

By its oral face the macronucleus sometimes reaches the level of the pectinellar ring, but in most cases it does not. Its average dimensions are 22 by 14 μ . The micronucleus is situated to one side above the oral face of the macronucleus (Fig. 3D).

Taxonomic affinities. Besides the differences mentioned in the foregoing species it differs from *A. campanulata* by having less concentric annuli on the oral half of the body; from the *A. phoxini* it differs by having more concentric annuli, somewhat thinner peristomial lips and by the absence of the protruding ledge of the pectinellar kinety and also by a different body shape. There is also no evidence of identity of this species with any of the species in SHULMAN's list (1962) or with any earlier description. Therefore we propose to designate this species *Apiosoma gobionis* n.sp.

Epistylis lwoffii Fauré—Fremiet, 1943

syn. *Rhabdostyla cyprini* Šrámek—Hušek, 1953, *Epistylis lwoffii*? Lom and Vávra, 1961

Lives on the body surface of *Cyprinus carpio*, *Abramis brama*, *Blicca björkna*, *Carassius carassius*, *Rutilus rutilus*, *Scardinius erythrophthalmus*, *Perca fluviatilis*, *Acerina cernua*, *Nemachilus barbatulus*, *Salmo trutta* m. *fario*, *Gobio gobio*, and occasionally on *Cottus gobio*. Fauré—Fremiet described this species from *Gasterosteus aculeatus*. The occurrence on other species of our fishes is possible.

This species occurs on different host species in ecoforms, differing each from the other chiefly by the shape of their stalk. FAURÉ—FREMIE (1943b) described populations from *Gasterosteus aculeatus*, consisting of small colonies comprising up to 8 individuals to one stalk. The colonies are never fixed directly to the host's skin, but the end of their stalk is shaped like a ring, embracing the scopolar end of *G. piscicola* and thus anchored to the fish.

LOM and VÁVRA (1961) described another ecoform from the surface of *Perca fluviatilis*. Only one part of the animal is attached to *Apiosoma*, the rest being fixed by fork-like or simple endings of their stalks directly to the skin. Often the ciliates form pseudocolonies on interweaving their stalks.

On the surface of *Cyprinus carpio*, *Carassius carassius*, *Abramis brama*, *Rutilus rutilus*, *Scardinius*

erythrophthalmus and *Salmo trutta* a third ecoform of this species occurs quite frequently. Here the colonies consist of up to 15 zooids, and the peduncle ends like an irregular plate slightly attached to the host's epithelium. The colonies and the body are shown in Pl. IV, Fig. 4 and 5.

The length of the grown-up zooids is about 80μ ; the greatest width is in the oral third of the body, the average width being about 33μ . The scopula is about 8μ broad, the stalk about $5-5.5 \mu$. The peristomial lip is well developed, its height amounting to 8.5μ on the infundibular and 4.8μ on the opposite side. The

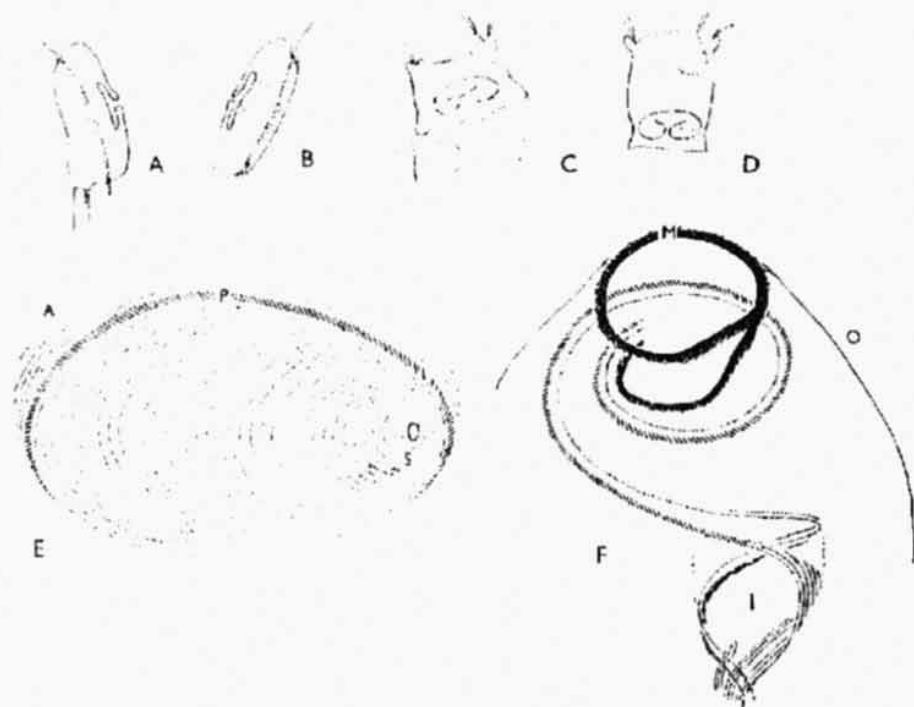


Fig. 4. A—D: part of the developmental cycle of *Epistylis leoffi*. A—zooid becomes a telotroch, scopula shifts eccentrically. B—telotroch, C—recently fixed ciliate, adhering to the substrate with whole aboral surface, D—a more advanced stage, which will start soon the stalk formation. E—aboral face of the newly formed telotroch as revealed by Klein's method. s—scopula, shifted eccentrically, p—pectinellar polykinety, a—concentric argentophilous annuli of the oral half of the body. F—buccal infraciliature of *E. leoffi* as revealed by protargol impregnation. M—circular myofibril, o—outline of the body, p—outer peristome, i—infundibulum. The scheme of ciliation is the same as in Fig. 3A.

peristomial lip is slightly vaulted, and is elevated some 5μ above the mouth of the infundibulum. The buccal ciliary apparatus follows the common peritrichean scheme (Fig. 4F). The circular myofibril within the peristomial lip, closing the peristomial space in contracted ciliates, has an inner branch supporting the outer end of the adoral zone.

The pellicle bears fine circular annuli, which are well demonstrated by the dry silver method after Klein together with the irregularly distributed openings of muciferous pores in form of heavily impregnated dots. Very often, the argentophilic annuli are bifurcated. The numbers of annuli are as variable as the numbers of the kinetics in most groups of ciliates. For instance, in populations from carps it varies from 41 to 54 in the anterior half of the body and from 39 to 44 in the

posterior aboral half; in trout populations from 50 to 57 and 44 to 52; on *Gobio* 46
gobio from 44 to 49 and 43 to 48.

The horse-shoed macronucleus is situated orally, below the peristome at approximately the same level as the contractile vacuole. The interphasic micronucleus is spherical, of 2—3 μ in diameter, situated closely to the macronucleus. Only in young, recently attached ciliates of the scyphidia—like stage, the horse-shoe of the macronucleus is shorter and thinner and is situated aborally, above the basal surface of the young ciliate.

The fibrillar system is weakly developed; protargol impregnation reveals only short stretches of longitudinal myofibrils, extending orally from the pectinellar fringe.

The cycle of the species is quite simple. In overcrowded populations or in unfavourable conditions, loaf-shaped telotrochs are formed. So far very little attention has been paid to these changes from the trophic ciliate to the swarmer. When the zooid starts to transform itself into the telotroch, the scopula begins to migrate to one side of the animal, just closely to the pectinellar kinety, so that the developing swarmer is attached obliquely to the stalk—the level of the pectinellar kinety almost coinciding with the longitudinal axis of the stalk. This shift is connected with a drastic change of the argentophile pattern of the aboral half of the animal (Fig. 4F); at one side the annuli turn into a cluster of argentophilic dots, at the opposite end they are double-spaced.

After the re-attachment of the swarmer to the substrate, it adheres to it at first with a large part of its aboral surface (Fig. 4C and Pl. IV). At this moment, the morphology of the oral part of the body soon becomes almost normal, but the scopula is not yet formed, the pellicle adhering to the substrate is full of argentophile dots (not all of them are, however, scopular kinetosomes). The number of argentophilic annuli between the pectinellar girdle and the border of the adhering surface is greatly reduced. Afterwards the ciliate gradually begins to assume the elongated shape of the grown-up ciliate, while still attached to the surface by the flattened aboral pole of the body, which secretes an irregular, flat basal platelet of the future stalk. At this stage the ciliates in most cases divide into two (or more) individuals. Then their aboral ends begin to take up the "adult" form and produce the stalk. That is why we find two up to several stalks attached to one flat, irregularly secreted basis (Plate IV, Fig. 2, 3) by which the thus formed colonies are anchored to the skin of the fish. The basal plate may also serve as a place of attachment for other telotrochs. The branching of the stalk is irregularly dichotomic. This cycle is characteristic for the third ecoform.

In spite of the large ciliate material observed, we have never found a cyst formation or induced it experimentally. There is also a common association of *E. hoeffi* of this ecoform with *Apiosoma*, but the mode of attachment is inverted to what is found on *Gasterosteus aculeatus* or perches. The flat irregular platelet, secreted by the settled swarmer, serves as a substrate for *Apiosoma*. Curiously enough, in the same population, where most of the *Apiosoma* were attached to basal platelets

17 of *Epistylis*, we found in turn, a certain number of *Epistylis lwoffii*, which were attached by tiny basal plates directly to the surface of the aboral part of the body of *Apiosoma*, most often in the aboral third of the body. However, *Epistylis* living in such an association can be observed in "colonies" of only two to three ciliates with short stalks.

FAURÉ-FREMIET did not observe the way in which *E. lwoffii* forms its anchoring annules round the caudal part of *Apiosoma*. Neither could we discover the mode in which the scopula of the settled swarmer produces the annular or fork-shaped endings of the stalk in *Epistylis* populations from the skin of perches.



Fig. 5. *Epistylis lwoffii* from the skin of *Acerina cernua*.
A - general appearance of a typical specimen.
B, C - other frequent shapes of stalk.

On the skin of *Acerina cernua* we observed populations of more stubby, solitary ciliates with very short stalks, ending by a flat disc attached to epithelial cells (Fig. 5; Pl. IV, Fig. 6, 7). For the most part they are not exceeding a body size of 25 by 40 μ . By their short unbranched stalks they could be members of the genus *Rhabdostyla*. A more detailed scrutiny reveals their appartenance to *Epistylis lwoffii* - they have the same structure of the buccal ciliary apparatus, arrangement of circumperistomial myofibril, the same external appearance of peristomial lips and epistomial disc as the young *Epistylis lwoffii*. In agreement are also the position of the macronucleus, the contractile vacuole and the number of argentophilic annuli. On some of the hosts we find a gradual transition between these specimens and those having an appearance typical for *E. lwoffii*, which may also form small colonies and where the stalk has one annular ending. Such aberrant populations on *Acerina* make the impression of a young *E. lwoffii* whose development was stopped as soon as it began to form the stalk. It may be also considered as a special ecoform, in which the coloniality is in regression (or not yet developed?). These populations also frequently harbour *Endosphaera engelmani*.

Another indication of a great plasticity of *E. lwoffii* is the occurrence of populations with considerably thinner stalks ($\frac{2}{3}$ of the normal width) and mostly unbranched, on the skin of *Nemachilus barbatulus*. On the gills of the same host we may find ciliates of the same habitus as on other fishes.

I. Relation of sessiline peritrichs to their hosts

This relation varies within the limits of simple ectocommensalism with a trend towards parasitism in cases of heavy invasions, although this tendency is much less pronounced than in trichodinids. Fish sessiline peritrichs may be given the commonly used term epizooites, more suitable than the old term of DEEGENER "symphoriontes", excluding parasitism. For their occurrence on the surface of fishes it may be more suitable to use the term "epifaunation" (BEERS 1962) than "infection" or "invasion".

Sessiline peritrichs—the same as the free living peritrichs, feed on bacteria and water-dispersed particles, thus being essentially host-independent in their food requirements, although they may, of course, ingest particles of detritus from the host's surface. Accordingly, their oral ends are mostly not hidden between the gill filaments, or in the scale- or skin interspaces, but are turned towards the surrounding water, which is well visible especially in *E. lwoffii*. THOMPSON et al. (1947) found a direct relation between the high content of bacteria in water and the abundancy of *Scyphidia ameiuri* on the surface of the fishes both in nature and in fishes brought to the laboratory. These observations were corroborated by our own.

However, these ciliates are dependent on the host as for living space, and the host exhibits a direct influence on them. This can be seen not only in the suitability of the host's surface for a given ciliate—here the objection could be made, that it is the ciliate whose telotroch chooses its host according to its requirements—but also in the seasonal variations of the ciliate numbers on the fish.

We found most numerous populations of *Epistylis lwoffii* and *Apiosoma* of all four species during the winter months—January, February, March, April—up to about middle of May. After that time, in spring, when the temperature raises and the fishes reassume their activity and begin to feed, and then during summer and autumn the epizooites are very rare or seem to be completely absent. Evidently, the surface of the fish becomes less suitable for the ciliates. This may be connected with some physiological changes of the host during the vegetative period. In autumn, when the temperature of the water is approximatively the same as in April or May, and the number of bacteria and organic particles dispersed in the water seems to be at least the same as in the early spring months, we find epizoa to be rare or absent. This is certainly due to the still "active" condition of the host. However, by which means—or metabolic products—this influence is brought about, is a matter of speculation.

A direct pathological action of these epizooites was reported by SURBER (1940): fingerlings of the largemouthed bass, two inches in length, died from large numbers of *Ambiphrya thaliformis* on their skin, unless they were treated by 1.5 % NaCl. SCHÄPERCLAUS (1954) observed also massive occurrence of *Apiosoma* on carps

49 without attributing to them a really injuring significance. FIJAN (1962) described losses on two months old carp fry caused by massive epifaunation by *Apiosoma*, which according to his microphotographs was a typical *A. piscicola*. Being unable to state any pathological changes on the gills or on the skin, he claimed that the ciliates prevented the normal oxygen and metabolic product diffusion on the gills and the skin. Without experimental evidence it is not easy to decide whether this is true or whether the losses were caused by a sort of irritation or allergy due to the great number of protozoa attached to the surface of the fish. As mentioned earlier, the scopula adheres to the epithelial cells very firmly and during massive epifaunation such irritation could be important, even without disintegrated epithelium of the skin.

II. Mutual relation of the species forming the faunule on the skin of the fish

There seems to be no pronounced relations to trichodinids although it was not examined experimentally in the same way as NOBLE'S studies with trichodinids and monogeous (1963). However epizoa disappear whenever trichodinids are present in massive populations. Interesting is the mutual association of *Epistylis lwoffii* with any of the four *Apiosoma* species described in this paper, which it finds on the skin of the occupied host. This association— as well as that of *Apiosoma* and *Erastophrya chattoni*— is in most cases aimed at the most useful exploitation of the attachment capacity of one of them. The ciliates always prefer the more advantageous way of attachment to the host's skin. For peritrichous epizooites the surface of the fish is a far less suitable substrate to settle on, because e.g., the carapax of the crustaceans— the mucous surface layers of epidermal cells with frequent desquamation of the cells — is not a very solid support. Therefore *Apiosoma* looks for the basal platelet— the single solid point on the mucous surface — of *Epistylis* populations on carps and vice versa *E. lwoffii* fastens itself to *Apiosoma* which has a large scopula even at the adult stage because on *Gasterosteus aculeatus*, *E. lwoffii* is for reasons unknown not capable to produce such a plate.

In general, *Apiosoma* is better adapted for life on such an unsolid surface as the skin of the fish. For this reason we know a great number of fish inhabiting species of *Apiosoma* but only few species of stalk-forming peritrichs. This adaptation— no stalk, but a scopula stretched into a large basal adhering surface— explains well the distribution of the family *Scyphidiidae* on water animals with a soft surface.

The association between *Apiosoma* and *E. lwoffii* is not unique. NENNINGER (1950) recorded a similar phenomenon from leeches: *Scyphidia hirudineorum* is attached to the basal platelet of the stalk of *Epistylis bimarginata*. Both instances have in common the mucous and unsolid skin of the fish and leech.

The predators of *E. lwoffii* are *Endosphaera engelmanni* which invades this

species as well as trichodinids, and *Hypocoma parasitica* Grüber. The latter species, known hitherto only from free-living peritrichs, agrees in its morphology entirely with the description of CHATTON and LWOFF (1950). 50

III. Cyst formation

In none of the species recorded any cyst formation could be observed. Only once we noticed a rounded cyst in a smear of *E. lwoffii*—which may have belonged to another species. The lack of cysts is a feature common with epizoites and trichodinids. Their distribution is perhaps sufficiently assured by telotrochs, the protection against unfavourable ecological conditions being entrusted to the host. Sessilina from the surface of amphibian larval stages produce cysts (e.g., *Rhabdostyla scyphidiformis* Vávra, 1960) which probably are the germ of new populations for amphibian larvae in the next year.

The great plasticity of *E. lwoffii* (ecoforms on different hosts) may lead to phylogenetic speculations, but we should only like to mention its meaning to the practical determinative viewpoint. It stresses the importance of careful morphological analysis before giving a specific rank to aberrant habitus of populations belonging to one species. Let us once more emphasize the features which we consider necessary to be included in a reliable description of the sessiline peritrichs of fishes:

1. Observation of living ciliates is absolutely necessary.
2. Shape of the body, morphology of the peristomial region. Attention should be paid to the great variability as well as to the stages of the developmental cycle. The body of peritrichous ciliates is uniform, so that all peculiarities will be helpful in the determination. Good drawings are necessary, photographs most advisable.
3. Size of the body; significant are the maximal values attained by the ciliates. Not important is the relation of the size of the body to the size of the macronucleus.
4. Morphology of the scopula or stalk.
5. Position of the micronucleus in relation to the macronucleus, their size.
6. Number of argentophilic annuli. Impregnation according to Klein's method most advisable; the annuli can be seen also in material fixed in sublimate-alcohol and observed under the phase-contrast microscope.
7. Host and localisation on the body.

Description of all additional features (buccal ciliature etc.) is most useful, but it is useless to mention the features common to all peritrichs in general.

Genus *Apiosoma* Blanchard, 18851. *Apiosoma piscicola* Blanchard, 1885

syn. *Glossatella piscicola* (Blanchard, 1885) Kahl, 1935.

Glossatella cyprini Šrámek—Hušek, 1953.

Glossatella cylindriciformis Chen, 1956 from the surface of Chinese pond fishes *Ctenopharyngodon idella*, *Mylopharyngodon piceus*, *Pseudaspius leptocephalus*, *Hypophthalmichthys molitrix*; is identical with *A. piscicola*.

A well characterised species, discussed sufficiently in the foregoing paragraphs.

2. *Apiosoma amoeba* (Grenfell, 1887) emend.

syn. *Scyphidia amoeba* Grenfell, 1887.

Glossatella amoeba (Grenfell, 1887) Kahl, 1935.

On the surface of *Gasterosteus aculeatus*.—The original description is insufficient. The amoeboid projections of the scopula are probably attached remnants of epithelial cells. ROUX (1901) determined a species he found on the fins of *Rutilus rutilus* as *Scyphidia amoeba*; however, except for the extensions of the scopula (probably of a character mentioned above) there is no evident proof of an identity of both species. According to ROUX's superficial description, he might have dealt with *A. piscicola*. The taxon was saved by TIMOFEEV (1962) who determined the ciliates found by him on *Gasterosteus aculeatus* as *A. amoeba*. PRECHT's *Scyphidia gasterostei* (1935) may be identical with this species, but the description is not complete. *A. amoeba* is a species requiring revision.

3. *Apiosoma cotti* (Voigt, 1902) emend.

syn. *Glossatella cotti* (Voigt, 1902) Kahl, 1935.

On *Gobio gobio* in Germany.—Described originally by VOIGT as ecological variety of *Glossatella tintinnabulum* (Kent, 1882). Bütschli, 1889, and not found since.—It has to be reexamined.

4. *Apiosoma glabra* (Roth, 1909) emend.

syn. *Cordylosoma glabra* Roth, 1909.

Glossatella glabra (Roth, 1909) Kahl, 1935.

Originally described from the skin of "fishes". KAHL expressed his doubt on the reliability of this insufficiently described species. However, in 1950 NENNINGER briefly recorded it from *Molge cristatus*.—Redescription necessary, if possible at all.

5. *Apiosoma micropteri* (Surber, 1940) emend.

syn. *Scyphidia micropteri* Surber, 1940.

On the surface of *Huro salmoides* and *Micropterus dolomieu*, USA.

6. *Apiosoma* sp. Raabe, 1952

syn. *Glossatella* sp. Raabe, 1952.

On the surface of *Esox lucius*.—Unidentified species.

7. *Apiosoma piriformis* (Tripathi, 1954) emend.

syn. *Glossatella piriformis* Tripathi, 1954.

On the surface of *Laboe rohita*, *Calta calta*, *Cirrhina surigata* and *C. reba*.—*Nomen dubium*, the description being quite insufficient.

8. *Apiosoma minuta* (Chen, 1961) emend.

syn. *Glossatella minuta* Chen, 1961.

Body surface of *Squaliobarbus curriculus*, *Parabramis pekinensis*, *Hemiculter leucisculus*, *H. eigen-*

mani, *Cyprinus carpio haematopterus*, *Alburnus alburnus*, *Pseudobagrus fulvidraco*, *Misgurnus anguillicaudatus* from the river Amur, Asia. — Reexamination of living specimens necessary. 52

9. *Apiosoma doliaris* (Timofeev, 1962) emend.

syn. *Glossatella doliaris* Timofeev, 1962.

Nasal cavity of *Scardinius erythrophthalmus* from the Neva Bay, USSR. Reexamination of living specimens necessary.

10. *Apiosoma megamicronucleata* (Timofeev, 1962) emend.

syn. *Glossatella megamicronucleata* Timofeev, 1962.

On the body surface of *Silurus glanis* in the Baikal Sea and river Amur, Asia. — Reexamination of living specimens necessary.

11. *Apiosoma nasalis* (Timofeev, 1962) emend.

syn. *Glossatella nasalis* Timofeev, 1962.

Nasal cavity of *Leuciscus walekii*, *Sarcochilichthys sinensis lacustris*, *Chilogobio czerskii*, *Acanthorhodeus asmussi*, *Carassius auratus gibelio*, river Amur, Asia. — Reexamination of living specimens necessary.

12. *Apiosoma conica* (Timofeev, 1962) emend.

syn. *Glossatella conica* Timofeev, 1962.

Not sufficiently differentiated from other species, especially from *A. amoeba*.

13. *Apiosoma campanulata* (Timofeev, 1962) emend.

syn. *Glossatella campanulata* Timofeev, 1962.

Body surface of *Perca fluviatilis*, *Lucioperca lucioperca*, rarely *Acerina cernua*, *Rutilus rutilus*, Europe. — Well characterised species.

14. *Apiosoma basiramosa* (Timofeev, 1962) emend.

syn. *Glossatella basiramosa* Timofeev, 1962.

Gills of *Opsariichthys unciostriis amurensis*, river Amur, Asia. Well characterised species.

15. *Apiosoma poteriformis* (Timofeev, 1962) emend.

syn. *Glossatella poteriformis* Timofeev, 1962.

Body surface of *Nemachilus barbatulus*, Neva Bay, USSR.

16. *Apiosoma robusta* (Zhukov, 1962) emend.

syn. *Glossatella robusta* Zhukov, 1962.

On the gills of *Dalia pectorails*. Rivers emptying into the Behring Sea. — Reexamination of living specimens necessary.

17. *Apiosoma peculiariformis* (Zhukov, 1962) emend.

syn. *Glossatella peculiariformis* Zhukov, 1962.

Rivers emptying into the Behring Sea. Insufficiently described.

18. *Apiosoma dallii* (Zhukov, 1962) emend.

syn. *Glossatella dallii* Zhukov, 1962.

Body surface of *Dalia pectoralis* in rivers emptying into the Behring Sea. Insufficiently described.

19. *Apiosoma gigantea* (Kandilov, 1964) emend.

syn. *Glossatella gigantea* Kandilov, 1964.

20. *Apiosoma phoxini* sp.n.

21. *Apiosoma gobionis* sp.n.

53 Genus *Scyphidia* Dujardin, 1841

1. *Scyphidia scorpaenae* Fabre—Domergue, 1888.

Body surface of *Scorpaena*, Atlantic. Nomen dubium—insufficiently described.

2. *Scyphidia acanthoclini* Laird, 1953.

On the gills of marine fish *Acanthoclinus quadridactylus*, New Zealand coast. Well characterized species.

3. *Scyphidia arctica* Zhukov, 1962

Gills of arctic marine fish *Liparis gilleus* and *Melletes papilio*.

Genus *Ambiphrya* Raabe, 1952

1. *Ambiphrya tholiformis* (Surber, 1943) Raabe, 1952.

On the body surface of *Huro salmoides* and *Micropterus dolomieu*, USA.

2. *Ambiphrya macropodia* (Davis, 1947) Raabe, 1952.

Surface of *Ameiurus nebulosus*, USA. Well characterized species.

3. *Ambiphrya miri* Raabe, 1952.

Body surface of *Nerophis ophidion*, The Baltic Sea. Well characterized species.

Genus *Calliperia* Laird, 1953

1. *Calliperia longipes* Laird, 1953.

Gills of *Oliverichthys melobesia*, *Ericentrus rubrus*, marine fish from the New Zealand coast. Well characterized species.

2. *Calliperia brevipes* Laird, 1953.

Gills of *Raja erinacea*, The Atlantic coast of Canada. Well characterized species.

Genus *Epistylis* Ehrenberg, 1838

1. *Epistylis gasterostei* Fauré—Fremiet, 1905.

A rather enigmatic ciliate. Described by a brief mention originally from *Gasterosteus aculeatus*, without any drawing. This old description gives no real idea about the ciliate's morphology. Therefore PRECHT (1935) determined as *Epistylis gasterostei* a species of *Glossatella* which he found on three spined sticklebacks and he rectified the name to *Scyphidia* (!) *gasterostei*. NENNINGER (1950) mentioned the occurrence of *Epistylis gasterostei* on *G. aculeatus* without any description. In 1943, FAURÉ—FREMIEU explains: "*E. gasterostei* ... dont la structure est celle d'une *Glossatelle* mais dont la base notablement élargie sécrète au niveau de la scopula un tube pédonculaire largement fixé sur le tegument ... de l'hôte".

A redescription is urgently needed.

2. *Epistylis hoeffii* Fauré—Fremiet, 1943.

Discussed above.

Genus *Zoothamnium* (Bory 1824) Ehrenberg, 1838, Stein, 1854

1. *Zoothamnium* sp. Jones and Job, 1938.

On the mouth epithelium of *Acentrogobius neilly* from India.

2. *Zoothamnium duplicatum* Kahl, 1933.

Found by PRECHT (1935) on bases of pectoral fins of *Cottus scorpius*, and solitarily on the proboscis of *Syngnathus typhle*.

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EXPLICATIONS TO PLATES

Plate I

Fig. 1. *Apiosoma piscicola* attached by its scopula to the basal plate of the stalk of a two-zooid colony of *Epistylis loeffli*, which is fixed on epithelial cells. The line indicates 40 μ .

Fig. 2. Detail of the basal part of *A. piscicola* associated with *E. loeffli* in the same way as in the preceding picture.

Fig. 3. Oil immersion lens picture of silver impregnated (Klein's dry method) aboral half of *A. piscicola*. s—scopula, I—concentric impregnable annuli, K—pectinellar kinety, M—impregnable muciferous pellicular pores.

Fig. 4. Scopula of *A. piscicola* attached to the basal plate (P) of the stalk of *E. loeffli*. Note the scopular cilia (arrow). The line indicates 10 μ .

Plate II

Fig. 1. Oil immersion lens picture of silver impregnated (Klein's method) *Apiosoma campanulata* with contracted peristome. Note the impregnable argentophilic annuli and muciferous pores.

Fig. 2. Contracted specimen of *A. campanulata*. Note the coarse striation of the pellicle. Arrow points to the peristomial space. The line indicates 20 μ . M—triangular macronucleus.

Fig. 3. *A. campanulata* in position on the skin epithelium of perch. Line indicates 20 μ .

Figs. 4 and 5. Different shapes of stubby specimens of *Apiosoma gobii* attached with their broad scopula to skin epithelium of *Gobio gobio*. Note the transversely oval shape of macronucleus (M). The line indicates 40 μ .

Fig. 6. More elongated specimen of *A. gobionis*.

Fig. 7. Elongated specimen of *A. gobionis*. As in preceding figure, the line indicates 20 μ .

Plate III

Fig. 1. *Apiosoma phoxini* (bottom) associated with *Epistylis loeffli* (top right) and *Erastophrya*

chattoni (top left). Annular stalks of *Erastophrya* and *Epistylis*, encircling the aboral end of *Apiosoma* 56
are not in the focus. The line indicates $40\ \mu$ in Figs. 1 to 4.

Fig. 2. *A. phoxini* with semi-contracted peristome; a *Tripartiella* glides upon its surface.

Fig. 3. Typical *A. phoxini* in feeding position. Arrow indicates the ridge, bearing the pectinellar kinety. M—macronucleus.

Fig. 4. A specimen with very broad aboral half of the body.

Plate IV

Figs. 1 to 5. *Epistylis lwoffii* from the skin of *Abramis brama* and *Salmo trutta m. fario*.

Fig. 1. Young ciliates fixed to epithelial cells with their broad scopula. The line indicates $20\ \mu$.

Fig. 2. A basal plate (P) anchoring stalks of several colonies of *E. lwoffii*, torn off the epithelial cells, the remnants of which are seen on its bottom side. The line indicates $20\ \mu$.

Fig. 3. A basal plate still fixed to the host's skin. The line indicates $20\ \mu$.

Fig. 4. Typical zooids of *E. lwoffii*. Line indicates $50\ \mu$.

Fig. 5. A colony of *E. lwoffii*. Line indicates $50\ \mu$.

Figs. 6 and 7. *E. lwoffii* from the skin of *Acerina cernua*. 44 a specimen with semicontracted peristome. F—the short stalk.

Fig. 7. The short stalk of another specimen. The line indicates $20\ \mu$.



