Heteropolaria colisarum Foissner & Schubert, 1977 (Protozoa : Epistylididae) of North American freshwater fishes

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Abstract. The peritrichous ciliate 'Epistylis sp.' involved in the 'red-sore' disease of various North American freshwater fishes is identified as Heteropolaria colisarum Foissner & Schubert, 1977. Its morphology is redescribed from observations of living and silver-stained organisms and biometrically analysed individuals. Extended zooids can be easily identified by their highly characteristic (elongated) body. Silver impregnations show a very high number of silverlines and a band-like, irregularly formed macronucleus. The genus Heteropolaria belongs to the family Epistylididae and is characterized by the eccentric location of the scopula of the swarmer and a peculiar myoneme in the peristomal disc that branches off the myoneme of the peristomal collar. Autochthonous occurrences of this species are known only from North America. In Europe only the related species H. lwoffi has been found. Histological sections give some evidence that the terminal platelet of the stalk is embedded in the dermal epithelium. This suggests that H. colisarum could be a parasite rather than an epizoon and that the often associated bacterium Aeromonas hydrophila is a secondary invader. However, further studies are necessary. Experiments showed a high infectivity of the ciliate and no host specificity. These characteristics are supported by the observations of other researchers. Preliminary laboratory tests showed that a single treatment with salt solution (NaCl) at a rate of 1.5% for 3h controlled H. colisarum.

Introduction

Since 1970 there has been a growing list of reports about a 'red-sore' disease of various North American freshwater fishes (Rogers 1971; Hubert & Warner 1975; Esch, Hazen, Dimock & Gibbons 1976; Miller & Chapman 1976; Hazen, Raker, Esch & Fliermans 1978). From the published figures and photomicrographs it is obvious that a ciliated protozoan belonging to the peritrichous family Epistylididae is always involved. As Rogers (1971) stated, the main problem with this *Epistylis* infestation is not mortality of fish but rejection by anglers because of the diseased appearance of the fish. But Miller & Chapman (1976) reported that on Badin Lake, North Carolina about 37 500 infested fish died over a period of about 13 days, Although this disease causes serious economic losses, no accurate description or identification of the epistylid involved has been published.

Without knowledge of the fish-parasitological reports cited above, Foissner & Schubert (1977) published a taxonomic description of a new epistylid, *Heteropolaria*

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colisarum, from the skin of the giant gourami, *Colisa fasciata* (Bloch). When we compared this species with the 'red-sore' epistylid it seemed likely that it was identical. To investigate this likelihood, we performed a taxonomic study on the American epistylid and tried to collect data about its infectivity, host specificity, treatment and management.

Materials and methods

Collection of samples

In 1974, one of the authors (G.L.H.) cursorily examined two green sunfish, *Lepomis cyanellus* Rafinesque, 12 cm long, that had visible whitish colonies of an epistylid on the dorsal fins. These fish had been kept in a concrete tank in the wet laboratory of the Fish Farming Experimental Station, Stuttgart, Arkansas for several months at about 21°C. No attempt was made at that time to identify the epistylid further.

In February 1981 an epizootic of an epistylid, later identified as *Heteropolaria* colisarum, occurred on green sunfish being cultured in a tank at the Fish Farming Experimental Station. (In retrospect we believe that the 1974 epistylid was also *H.* colisarum.) About 10% of the 170 fish (9–11 cm long) in a 264-1 circular tank were visibly infested. The temperature was maintained at 29°C. The water inflow was 1 gal/min (=about one exchange per hour), total alkalinity 275 ppm, and pH7.6; adequate aeration was supplied. However, because of the rather high density of fish, the water contained a moderate amount of organic material, and it appeared that the sporadic mortality was caused by secondary bacterial infection of the lesions. Colonies were clearly visible to the naked eye as white masses on the tips of the fins, particularly the dorsal fin. Many of the scales bore smaller inconspicuous colonies.

Host	Date	Locality	Type of water	Tempera- ture (°C)	Fish length (cm)	Prevalence
Channel catfish	1974	Unknown	Unknown	Unknown	Unknown	Unknown
(Ictalurus punctatus)	June 1978	Idaho	Raceway; geo- thermal water	25–28	9	High
	May 1979	Gallatin, Tennessee	Heated wastewater	24	10–14	High
	July 1981	Gallatin, Tennessee	Heated wastewater	29	20–25	High
	June 1981	Gallatin, Tennessee	Heated wastewater	26	20	High
	August 1982	Gallatin, Tennessee	Heated wastewater	25–28	8–9	High
Bluegill (Lepomis	Sept. 1979	South Carolina	Reservoir 6800 acres	25–27	13–17	1%
macrochirus)	Jan. 1982	California	San Joaquin River	24	14	Unknown
Green sunfish (Lepomis cyanellus)	Feb. 1981	^a Stuttgart, Arkansas	70 gallon tank rearing	29	6–11	10%

Table 1. Heteropolaria colisarum of North American freshwater fishes

^aU.S. Fish and Wildlife Service, Fish Farming Experimental Station.

During the period 1974–82, referral specimens originating from epistylid epizootics in channel catfish, *Ictalurus punctatus* (Rafinesque), from Idaho and Tennessee and bluegills, *Lepomis macrochirus* Rafinesque, from South Carolina and California (Table 1) were also cursorily examined by one of the authors (G.L.H.). More thorough study in 1982 showed all of them to be lesions of *Heteropolaria colisarum*.

Investigation of Heteropolaria colisarum

Living material. Anaesthetized or killed fish were examined under the dissection microscope at $\times 10$ for the presence of colonies on the fins. Simple wet mounts were made from material excised from lesions. Sketches and photographs were made of the most typical forms. Measurements were made at magnifications of $\times 63$ to $\times 400$ with an ocular micrometer. Photographs were made with an automatic Olympus photomicrographic unit.

Stained slides. Slides for dry silver impregnation were prepared from living material according to the method of Foissner (1976). Silver proteinate (protargol) staining following Bouin fixation was done according to the method of Foissner (1982). This rather complicated method is especially appropriate for revealing the infraciliature and other details such as the macronucleus and the myoneme system. Drawings were made with the aid of a camera lucida.

Histological examination of skin of fish

To determine whether the stalks of *H. colisarum* are anchored *on* the epithelial cells or *in* the skin, we investigated the skin of a heavily infested fish by standard histological techniques. Sections were stained with haematoxylin and eosin or Giemsa's.

Experiments

Infection experiment. Twenty infested green sunfish were placed in a 75-l aquarium and maintained for 13 months, after which smaller green sunfish (6–7 cm long) were added and the fish were divided among three aquaria. One was equipped with a heater to maintain a temperature of 22°C, near the temperature of the water in the donor epizootic; water temperatures in the other two fluctuated with seasonal temperature changes (14–26°C).

Host specificity experiment. Because H. colisarum was originally described from an Indian fish on a different continent (Europe) it was desirable to determine if host specificity was involved. We placed a fingerling channel catfish, goldfish, *Carassius auratus* (L.), and golden shiner, *Notemigonus crysoleucas* (Mitchill), in a basket in the contaminated aquarium after minor clipping of the dorsal fins to facilitate infection.

Treatment experiments. Laboratory experiments with salt solutions at various concentrations were performed. Tests were performed in 2-1 beakers aerated with air stones.

Results

Description of Heteropolaria colisarum Foissner & Schubert, 1977 from green sunfish (Lepomis cyanellus)

A detailed description of this species was given by Foissner & Schubert (1977). Thus, we

		Statistics							
No.	Character	$\dagger ar{X}$	М	sd	\$ _X	v	Range	n	
(1)	Number of silverlines from the oral	[‡] 184·3	185.0	16.7	3.3	9.1	125-210	25	
	apparatus to the aboral ciliary	[‡] 173.5	170.0	13.9	4.4	8.0	150-195	10	
	band	[‡] 194·1	195.0	8.6	2.2	4.4	180-206	15	
(-)	Number of silverlines from the aboral	132.5	135.0	9.4	1.9	7.1	110-150	25	
	ciliary band to the scopula	115.7	120.0	10.3	3.3	8.9	100-130	10	
		130.3	128.0	8.0	2.1	6.1	120-150	15	
(3)	Distance between two silverlines in	0.7	0.7	0.05	0.01	7.7	0.6-0.8	25	
	the middle part of the body	0.6	0.6	0.13	0.04	21.7	0.4-0.7	10	
	(μm)	0.4	0.4	0.11	0.03	25.7	0.2-0.7	15	
(4) Number of pell $100\mu\text{m}^2$	Number of pellicular pores per	18.2	18.0	4.2	0.8	23.2	10-30	25	
	$100\mu{\rm m}^2$	19.9	18.0	4.9	1.6	24.7	15-27	10	
		16.6	15.0	3.8	1.0	23.1	13–25	15	
(5) Length of	Length of macronucleus (μm)	92.8	90.0	12.4	2.5	13.4	75–115	25	
		121.1	120.0	12.7	4.0	10.5	100-140	10	
		81.8	84.0	10.7	2.1	13.0	70–110	25	
(6)	Width of macronucleus (μm)	8.7	8.4	1.0	0.2	11.9	7–11	25	
(0)		8.4	8.2	1.2	0.4	14.7	7–11	10	
		7.0	7.0	0.6	0.1	8.8	6-8	25	

Table 2. Biometrical characterization of Heteropolaria colisarum*

* Measurements of items 1 to 4 above based on dry silvered specimens, and items 5 and 6 on protargol stained material.

† Abbreviations: \bar{X} =arithmetic mean, M=median, sd=standard deviation, $s_{\bar{X}}$ =standard error of arithmetic mean, V=coefficient of variation, n=sample size.

‡For each of the six characteristics, the upper row of data refers to a population from Arkansas, U.S.A. (host, *Lepomis cyanellus*); the middle row refers to a population from Gallatin, Tennessee, U.S.A. (host, *Ictalurus punctatus*); and the lower row to a population from an aquarium in Stuttgart, Germany prepared from data published by Foissner & Schubert (1977) (host, *Colisa fasciata*).

describe here only the most prominent features and some new observations. Biometric data are given in Table 2.

Colony size. The smallest colonies observed were those on scales which were visible at a minimum of $\times 10$ magnification. The larger colonies on the fins tended eventually to cover the entire tips of the fins and reached a length of 3 cm on fish 9 to 11 cm long. They formed a very characteristic pulvinate whitish coat (Fig. 1).

Stalks. Colony stalks are as long as 1.18 mm (but usually shorter) and are 12 to $35 \mu \text{m}$ in diameter. Branching is dichotomous, ranging from no branches or one branch on very

Figure 1. Channel catfish with visible colonies (arrows) of Heteropolaria colisarum on dorsal and pectoral fins.

Figures 2 & 3. Photomicrographs of Heteropolaria colisarum.

Figure 2. Small colony with four slightly contracted zooids.

Figure 3. Part of a large colony with one fully extended zooid, showing the large vestibulum (V).



short stalks to many branches on long stalks (Figs 13–14). As many as six short branches, each with a zooid, occur at the tips of some stalks (Figs 2, 4, 5 & 12). The stalks of the younger zooids are about half the diameter of the parent stalk (Fig. 5). The terminal platelet (some workers have wrongly named this structure the 'scopula') of the stalk is about $33 \,\mu$ m in diameter and $12 \,\mu$ m thick in histological sections.

Morphology of zooid. Body form of extended zooids is cylindroid and elongated, never bell-shaped (Figs 3, 4 & 12). Extended specimens were 220 (150–300) um long and 47 (40-60) μ m wide (seven measurements). The base of the zooid is about half as wide as the peristomal collar. Contracted zooids are pyriform, less than half as long as the extended forms; e.g. a zooid that is $300 \times 60 \,\mu\text{m}$ extended becomes $120 \times 90 \,\mu\text{m}$ when fully contracted (Figs 5 & 12). In a living colony many individuals are often not fully contracted or extended (Figs 2 & 5). Such zooids lose their characteristic form; nevertheless, they can be easily misinterpreted as 'typical' forms. Unlike many other large epistylids, this species does not show an occlusion of the stalk by the posterior part of the body when it fully contracts. Many fine folds in the pellicle appear along the whole body. In an attempt to preserve normal (extended) forms, we tried nickel sulphate (about 1%) and hot water separately. Seven zooids in nickel sulphate were 200 (180–200) by 50 (45–60) μ m. Those killed in hot water and preserved in 10% formalin appeared normally extended. The pellicle is very finely transversely striated. Cytoplasm is whitish, densely filled with food vacuoles and large, clod-like inclusions 1 to 10μ m in diameter which show a characteristic funnel-shaped configuration in the posterior part of both the living and fixed zooid (Figs 3, 4 & 12). The contractile vacuole is at the dorsal side of the large, funnel-shaped vestibulum immediately below the pronounced peristomal collar. The macronucleus is band-like, without a regular form, often more or less coiled and twisted (Figs 12 & 15).

The peristomal disc is flat to hemispherical, sometimes umbilicated in the centre (Figs 2, 4 & 12). Haplokinety and polykinety describe about 1.5 turns (about 560°) at the peristomal disc before they enter the vestibulum, where they describe about two turns. The polykinety forms three peniculi in the lower half of the vestibulum. Peniculus 1 reaches the cytostome, and the very short peniculus 3 and the haplokinety end about 10μ m before the cytostome and consist of only two rows of basal bodies. Peniculus 2 is between the two others and ends about 20μ m before the cytostome. The germinal (stomatogenic) kinety extends to the middle of the vestibulum, where it passes

Figures 6 & 7. Heteropolaria colisarum, photomicrographs of protargol stained swarmer.

Figure 6. Aboral view showing scopula (S) and macronucleus (Ma).

Figure 7. Oral view part 1 of the myoneme system (My_1) , part 2 of the myoneme system (arrow) and haplokinety and polykinety (Hi+Pi).

Figures 4 & 5. Heteropolaria colisarum, photomicrographs of living zooids.

Figure 4. Part of a large colony with fully extended zooids showing peristomal collar (PC), vestibulum (V), primordium of the aboral ciliary band (PACB) and scopula (S).

Figure 5. Small colony with two slightly and one fully contracted zooid. At the right corner is part of a histological section showing two stalks of *H. colisarum* deeply embedded in the skin of a bluegill (arrows).





immediately beneath the haplokinety. It consists of irregularly distributed basal bodies (Figs 15–17).

The primordium of the aboral ciliary band, in the posterior third of the body, is marked by a ring-like bulge in living zooids and by two closely adjacent silverlines in stained materials (Figs 4 & 21). It consists of many short kineties (Fig. 15). The scopula, in the centre of the aboral pole, consists of many argyrophilic granules (basal bodies?) and is surrounded by a ring of larger, more regularly distributed granules (Fig. 15).

The strongly developed myoneme system consists of two parts: the rather thin myonemes, which extend from the haplokinety to the scopula (Figs 8 & 9, My₃), and a thick myoneme in the peristomal collar, from which a short branch extends into the peristomal disc (Figs 7 & 8, My₁, My₂).

The silverline system of the densely striped type consists of many ring-like silverlines that are connected by short, vertically oriented ones at irregular intervals. Some silverlines are bifurcated or end blindly in the pellicle; and some are so close together that they appear to be a single silverline. This phenomenon was called 'Pseudostreifung' by Foissner & Schubert (1977) and is caused by shrinkage of the zooids during dehydration. Many pellicular pores are closely attached to the silverlines (Figs 10, 11, 20 & 21).

Morphology of swarmer (telotroch). The swarmer is ellipsoidal, about 75 by $50\,\mu$ m, and flattened. The scopula is in the typical eccentric 'heteropolarid' position, but this characteristic is evident only in the fully developed swarmer (Figs 6 & 19). Developing swarmers show the expected transition stages in which the scopula moves from 'central' to more or less 'eccentric' positions. Kineties of the aboral ciliary band, which have about eight basal bodies each, bear long (about 20 μ m) cilia. All other argyrophilic structures are similar to those of normal, contracted zooids (Figs 6, 7, 18 & 19).

Histopathology

Heteropolaria colisarum occurs only on the skin, never on the gills. The histological sections give some evidence that the terminal platelet of the stalk is embedded in the dermal epithelium of the fins (Fig. 5). In most places the skin appears fairly normal but there are areas where the epithelium and some of the underlying dermis are extremely necrotic. These are undoubtedly the areas where secondary infections occur.

Figures 8–11. *Heteropolaria colisarum* after protargol silver impregnation (Figs 8 & 9) and dry silver impregnation (Figs 10 & 11).

Figure 8. Infraciliature and myoneme system in the oral part of an 'adult' in lateral view showing haplokinety (Hi), polykinety (Pi), germinal kinety (G), and part 3 of the myoneme system (My₃).

Figure 9. Infraciliature and myoneme system in the oral part of an 'adult' in ventral view showing the entrance to the vestibulum (V) where the myoneme system is formed like the arc of a gate (arrows), and the haplokinety and polykinety (Hi+Pi).

Figure 10. Part of the narrowly striated silverline system in the oral region showing destroyed outer part of the oral apparatus (OA) and pellicular pores (PP).

Figure 11. Part of the narrowly striated silverline system in the aboral region showing the scopula (S) and stalk (ST).



Infectivity, host specificity, treatment

The infection experiment showed infection of the new fish within 3 days. No exact measurements were made, but *Heteropolaria* infections were maintained at 14–29°C with very obvious colonies at 22–29°C.

No host specificity was found. *Heteropolaria colisarum* became visible in 1 to 6 days on all test species.

After the conclusion of five tests, salt at a concentration of 1.5% for 3 h was shown to be the most effective treatment. A solution of 1.5% for 2 h did not achieve complete control. Lower concentrations for longer periods (1% salt for 4 h) proved ineffective, and higher concentrations for shorter periods of time were either ineffective (2% salt for 1 h), or very toxic to the fish (2% for 2 h). Preliminary results clearly show that some fish cannot tolerate the salt treatment. Channel catfish of 9 g or larger tolerated it well (none died); green sunfish and goldfish tolerated it fairly well (2 out of 9 green sunfish and 3 out of 9 goldfish died) and golden shiners were unable to tolerate it (all died).

Discussion

Systematic position of Heteropolaria colisarum and comparison with related species

The most characteristic feature of the genus *Heteropolaria* is the eccentric location of the scopula of the swarmer (Foissner & Schubert 1977; Figs 6 & 19). Recently, Foissner (1983) added a second criterion, with some reservations, based on his studies of *H. lwoffi*: the peculiar myoneme of the peristomal disc that branches off the myoneme of the peristomal collar (Figs 7 & 18). In our opinion, these features justify the separation of these species from the genus *Epistylis* to which they would have formerly been referred.

Corliss (1979) placed the genus *Heteropolaria* into his poorly defined family Operculariidae. Guhl (1979), who overlooked *Heteropolaria* in his revision, furnished the Operculariidae and Epistylididae with new diagnoses, from which it is obvious that *Heteropolaria* belongs to the family Epistylididae because neither *H. colisarum* nor *H. lwoffi* has a stalked peristomal disc and both species have a well-developed peristomal collar. Rogers (1971), Hubert & Warner (1975), Esch *et al.* (1976), Miller & Chapman

Figures 12-14. Heteropolaria colisarum sketched from life.

Figure 12. A group of zooids showing one contracted (C).

Figures 13 & 14. Different forms of colonies.

Figures 15–17. Sketches from protargol silver impregnated specimens.

Figure 15. Infraciliature of an 'adult' in lateral view showing peristomal disc (PD), peristomal collar (PC), haplokinety (Hi), germinal kinety (G), polykinety (Pi) continuing within the infundibulum as P_1 and forming the peniculi (P_2 and P_3), the cytopharynx (Cy), macronucleus (Ma), primordium of the aboral ciliary band (PACB) and stalk (ST).

Figure 16. Higher magnification of the infraciliature of the oral apparatus in ventro-lateral view.

Figure 17. Detail of the infraciliature of the oral apparatus showing the germinal kinety (G), haplokinety (Hi) and peniculi 1 and 2 (P_1 , P_2).



(1976), and Hazen *et al.* (1978) also classified their species, which are probably identical with *H. colisarum*, as an *Epistylis* rather than an *Opercularia*. Only two species are similar to *H. colisarum*: *H. lwoffi* (Fauré-Fremiet, 1943) and *Epistylis longicorpora* Uyemura, 1938. *Heteropolaria lwoffi* differs from *H. colisarum* by its size (50–80 by 30–40 μ m), its pronounced bell-shaped body, its horseshoe-shaped macronucleus, its weak contractility, its short stalk, and the number of silverlines—about 90 (Lom 1966; Foissner 1983). *Epistylis longicorpora* resembles *H. colisarum* at first glance. However, it is larger (350–429 by 53–96 μ m) and has a very small horseshoe-shaped macronucleus. Contracted zooids show a snout which is never observed in contracted specimens of *H. colisarum*. Thus, we agree with Uyemura (1938) that *E. longicorpora* is closely related to the common *Epistylis plicatilis*.

Occurrence and host specificity

There is no significant difference between *H. colisarum* as originally described by Foissner & Schubert (1977) and our '*Epistylis*' from North American freshwater fishes. The biometrical values are also very similar (Table 2). Therefore, we are convinced that it is the same species. The characteristic body shape of *H. colisarum* also permits an identification with photomicrographs. Those published by Hubert & Warner (1975) and Miller & Chapman (1976) look so similar to ours that we have no doubt that they show *H. colisarum*. The *Epistylis* sp. of Rogers (1971), Esch *et al.* (1976) and Hazen *et al.* (1978) is very probably also *H. colisarum*. There are many reports about *Epistylis* sp. on fishes throughout the world (e.g. Fischthal 1953; Hutton 1964; Rauckis 1970; Dechtiar 1972; Miyazaki & Egusa 1973; Crites 1977; Hoffman 1978). Unfortunately, no accurate descriptions or photomicrographs of these *Epistylis* are available. Consequently, positive identification is impossible. The *Epistylis* sp. of Fischthal (1949) and Migała (1970) looks similar to *H. lwoffi*.

Heteropolaria colisarum was originally described from *Colisa fasciata*. These fish were imported from eastern Asia (Foissner & Schubert 1977). Thus no autochthonous occurrence is known from Europe, although there are many reports of *H. lwoffi* in Europe (e.g. Fauré-Fremiet 1943; Lom & Vávra 1961; Lom 1966; Lom, Golemansky & Grupchéva 1976; Scheubel 1973; Foissner 1983). There is no accurate report of this species from North America (Hoffman 1978), but the still growing list of reports of *H*.

Figures 18 & 19. Swarmer of Heteropolaria colisarum after protargol silver impregnation.

Figure 18. Oral view showing part of the myoneme system (My_1, My_2) .

Figure 19. Aboral view showing the scopula (S) and aboral ciliary band (ACB).

Figures 20 & 21. 'Adult' of Heteropolaria colisarum after dry silver impregnation.

Figure 20. Silverline system showing pseudoband (P) and primordium of the aboral ciliary band (PACB).

Figure 21. Higher magnification of the silverline system in the area of the primordium of the aboral ciliary band (PACB) showing vertically oriented silverlines (VS), horizontally oriented silverlines (HS), pellicular pores (PP), and a pseudoband (P).

colisarum show that this species is very common there. Rogers (1971) stated that cases of the disease have been reported from virtually every state in the south-eastern U.S. Other localities in North America were mentioned in the studies of Hubert & Warner (1975), Miller & Chapman (1976), Esch *et al.* (1976) and Hazen *et al.* (1978). These observations suggest that *H. lwoffi* is restricted to European fishes whereas *H. colisarum* prefers North American ones.

The studies of Foissner & Schubert (1977), Rogers (1971), Hubert & Warner (1975), Esch *et al.* (1976), Miller & Chapman (1976) and Hazen *et al.* (1978) proved that *H. colisarum* occurs on many kinds of fish belonging to different genera and families. Although not all species of fish are infested (Miller & Chapman 1976), we may conclude that host specificity in *H. colisarum* is either lacking or very weak. This view is supported by our infection experiment.

Heteropolaria colisarum: a parasite or an epizoite?

Heteropolaria colisarum is often associated with the Gram-negative bacterium, Aeromonas hydrophila (Esch et al. 1976; Miller & Chapman 1976; Hazen et al. 1978), but it is not clear whether *Heteropolaria* or *Aeromonas* is the primary invader. Rogers (1971) favours Heteropolaria, and Hazen et al. (1978) Aeromonas, but Miller & Chapman (1976) and Foissner & Schubert (1977) believe that Heteropolaria can be a primary invader as well as a secondary one. Hazen et al. (1978) especially prefer Aeromonas because they found Heteropolaria in only 35% of 114 lesions from 114 fish, whereas Aeromonas was present in 90% of the same lesions. However, they did not consider the possibility that the rather heavy colonies of H. colisarum could have fallen off the fish. They also stated that 'at no time was a stalk of *Epistylis* sp. observed to penetrate the epithelium covering the scales of bass'. On the contrary, the findings of Rogers (1971), as well as ours, give some evidence that *H. colisarum* could be a primary pathogen. The embedding of the stalk in the dermal epithelium (Fig. 5) causes the dermis to be inflamed and haemorrhagic (perhaps due to secondary bacterial infection!). This view is indirectly supported by the fact that the closely related species, H. lwoffi, which does not cause fish mortality (Lom 1966, 1973; Scheubel 1973) does not penetrate the epidermis (Lom 1973). However, more solid experimental research has to be done to enlighten the Epistylis-Aeromonas relationship.

Control of Heteropolaria colisarum

Epistylid infestations, particularly in the United States, have long been a formidable challenge for those seeking effective treatments. The most common treatment recommendations found in the literature have involved salt (NaCl) or salt combinations (Davis 1953; Reichenbach-Klinke 1966; Hubert & Warner 1975; Hoffman 1978; Rogers 1971; Fischthal 1949). Formalin, quinacrine hydrochloride, sulfaquine, and other chemicals have been tried with various degrees of success to control *Epistylis* sp. and *Zoothamnium* sp. on freshwater shrimps (Sindermann 1977; Lightner 1977; Roegge, Rutledge & Guest 1977). Hubert & Warner (1975) who tried several chemicals to control *H. colisarum*,

found that salt plus formalin and salt at 1.5% for 1 h were the most effective treatments. These results are similar to ours, except that we found a 3-h treatment to be necessary. Rogers (1971) suggested a 2.0% salt bath for 5 min, which was ineffective in our tests.

Esch *et al.* (1976) found positive correlations between thermal loading, body condition, and prevalence of *H. colisarum*. This correlation is supported by our results, which show very obvious colonies only at $22^{\circ}-29^{\circ}$ C. Because peritrichs feed on bacteria and colloidal particles (Schlick 1943; Foissner & Schubert 1977) that are numerous in nutrient-rich water, reduction of nutrients (organic overload) is advised for control in natural bodies of water, as well as in aquaculture.

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References

- Corliss J.O. (1979) The Ciliated Protozoa. Characterization, Classification and Guide to the Literature, 2nd edn. Pergamon Press, Oxford.
- Crites J.L. (1977) The occurrence of *Epistylis niagarae* (Ciliophora: Peritrichida) on fishes from the island region of western lake Erie. *Ohio Journal of Science* **77**, 193–194.
- Davis H.S. (1953) Culture and Diseases of Game Fishes. University of California Press, Berkeley.
- Dechtiar A.O. (1972) Parasites of fish from Lake of the Woods, Ontario. Journal of the Fisheries Research Board of Canada 29, 275–283.
- Esch G.W., Hazen T.C., Dimock R.V., Jr. & Gibbons J.W. (1976) Thermal effluent and the epizootiology of the ciliate *Epistylis* and the bacterium *Aeromonas* in association with centrarchid fish. *Transactions of the American Microscopical Society* **95**, 687–693.
- Fauré-Fremiet E. (1943) Commensalisme et adaptation chez une vorticellide: *Epistylis lwoffi* n.sp. *Bulletin de la Société Zoologique de France, Paris* 68, 154–157.
- Fischthal J.H. (1949) *Epistylis*, a peritrichous protozoan, on hatchery brook trout. *Progressive Fish-Culturist* 11, 122–124.
- Fischthal J.H. (1953) Parasites of northwest Wisconsin fishes IV. Summary and limnological relationships. *Transactions of the Wisconsin Academy of Sciences, Arts and Letters* **42**, 83–108.
- Foissner W. (1976) Erfahrungen mit einer trockenen Silberimprägnationsmethode zur Darstellung argyrophiler Strukturen bei Protisten. Verhandlungen der Zoologisch-Botanischen Gesellschaft in Wien 115, 68–79.
- Foissner W. (1982) Ökologie und Taxonomie der Hypotrichida (Protozoa: Ciliophora) einiger österreichischer Böden. Archiv für Protistenkunde 126, 19–143.
- Foissner W. (1983) Morphologie und Infraciliatur zweier ectocommensaler Ciliaten (Protozoa: Ciliophora) von Cyprinus carpio L. (Pisces: Cypriniformes): Heteropolaria lwoffi (Fauré-Fremiet, 1943) (Peritrichida: Epistylididae) und ihr Predator Pseudoamphileptus macrostoma (Chen, 1955) nov. gen. (Pleurostomatida: Amphileptidae). Zoologisches Jahrbuch Systematik 110, 399–418.
- Foissner W. & Schubert G. (1977) Morphologie der Zooide und Schwärmer von Heteropolaria colisarum gen. nov., spec. nov. (Ciliata, Peritrichida), einer symphorionten Epistylidae von Colisa fasciata (Anabantoidei, Belontiidae). Acta Protozoologica 16, 231–247.
- Guhl W. (1979) Beitrag zur Systematik, Biologie und Morphologie der Epistylidae (Ciliata, Peritricha). Archiv für Protistenkunde 121, 417-483.
- Hazen T.C., Raker M.L., Esch G.W. & Fliermans C.B. (1978) Ultrastructure of red-sore lesions on largemouth bass (*Micropterus salmoides*): association of the ciliate *Epistylis* sp. and the bacterium *Aeromonas hydrophila. Journal of Protozoology* 25, 351–355.

- Hoffman G.L. (1978) Ciliates of freshwater fishes. In: *Parasitic Protozoa*, Vol. II (ed. by J. P. Kreier), pp. 583-632. Academic Press, London.
- Hubert W.A. & Warner M.C. (1975) Control of *Epistylis* on channel catfish in raceways. *Journal of Wildlife Diseases* 11, 241–244.
- Hutton R.F. (1964) A second list of parasites from marine and coastal animals of Florida. *Transactions of the American Microscopical Society* 83, 439–447.
- Lightner D.V. (1977) Ciliate disease of shrimps. Developments in Aquaculture and Fisheries Science 6, 56-60.
- Lom J. (1966) Sessiline peritrichs from the surface of some freshwater fishes. Folia Parasitologica (Praha) 13, 36–56.
- Lom J. (1973) The mode of attachment and relation to the host in *Apiosoma piscicola* Blanchard and *Epistylis lwoffi* Fauré-Fremiet, ectocommensals of freshwater fish. *Folia Parasitologica (Praha)* **20**, 105–112.
- Lom J. & Vávra J. (1961) Epistylis lwoffi(?) from the skin of perches. Acta Societatis Zoologicae Bohemoslovenicae 25, 273-276.

*

- Lom J., Golemansky V. & Grupcheva G. (1976) Protozoan parasites of carp (*Cyprinus carpio L.*): a comparative study of their occurrence in Bulgaria and Czechoslovakia, with the description of *Trichodina perforata* sp. n. Folia Parasitologica (Praha) 23, 289–300.
- Migała K. (1970) [Investigation into external parasites of carp (Cyprinus carpio L.) under conditions of intensified pond production. Protozoa]. Roczniki Nauk Rolniczych 92, 49–72. (In Polish.)
- Miller R.W. & Chapman W.R. (1976) *Epistylis* and *Aeromonas hydrophila* infections in fishes from North Carolina reservoirs. *Progressive Fish-Culturist* **38**, 165–168.
- Miyazaki T. & Egusa S. (1973) [Ulcer disease of goldfish and crucian carp and *Epistylis* disease of colour carp.] *Fish Pathology* 7, 115–124. (In Japanese.)
- Rauckis E. (1970) [The seasonal changes in the pike parasitic fauna in the lake Dusia.] Acta Parasitologica Lituanica 10, 129–135. (In Russian.)

Reichenbach-Klinke H.H. (1966) Krankheiten und Schädigungen der Fische. G. Fischer Verlag, Stuttgart.

- Roegge M.A., Rutledge W.P. & Guest W.C. (1977) Chemical control of *Zoothamnium* sp. on larval *Macrobrachium acanthurus. Aquaculture* **12**, 137–140.
- Rogers W.A. (1971) Disease in fish due to the protozoan *Epistylis* (Ciliata: Peritricha) in the southeastern U.S. *Proceedings of 25th Annual Conference of the Southeastern Association of Game and Fish Commissioners* **25**, 493–496.
- Scheubel J. (1973) Die sessilen Ciliaten unserer Süsswasserfische unter besonderer Berücksichtigung der Gattung Apiosoma Blanchard. Zoologisches Jahrbuch Systematik 100, 1–63.
- Schlick W. (1943) Ernährt sich Carchesium polypinum von kolloiddispersen organischen Stoffen? Zoologischer Anzeiger 144, 59–72.
- Sindermann C.J. (1977) Ciliate infestation (*Epistylis*) of freshwater shrimps. *Developments in Aquaculture and Fisheries Science* **6**, 91–94.
- Uyemura M. (1938) On three new Epistylis from the fresh water mussel, Anodonta lauta v. martens. Annotationes Zoologicae Japonenses 17, 78–87.