

THE PROTOZOAN ASSOCIATES OF SOME CRUSTACEANS

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ABSTRACT

Protozoan associates found on the estuarine shrimp *Metapenaeus monoceros* (Fabricius) and the tanaidacean *Apeudes chilensis* (Chilton) from Cochin backwaters have been described. A peritrich ciliate *Zoothamnium rigidum* (Precht.), a heterotrich *Stentor coeruleus*, Ehrenberg and a microsporidean were recorded in both the hosts. Another ciliate *Lagenophrys cochinensis* (Santhakumari and Nair) was noticed only in *A. chilensis*. Experimental infestation of *Z. rigidum* on shrimp and the effect of stress produced by high turbidity and low oxygen tension in the infested tanaidacean were carried out. Most of the infested tanaidaceans were found dead, showing the potentially harmful nature of this ciliate.

INTRODUCTION

Crustaceans are known to harbour several species of ectocommensalic ciliates especially peritrichs (Kidder and Summers, 1935; Precht, 1935; Stiller, 1953; Debaiseux, 1959; Corliss and Brough, 1965; Fenchel, 1965 and Kinne, 1965). Many burrowing animals provide shelter for others as they happen to accommodate harmless "boarders" similar to those observed in *Chaetopterus* and sea urchin (Fisher and Mac Ginite, 1928). But the relationship between the peritrich ciliate *Zoothamnium* sp. and penaeid shrimps has shown a possible predilection to mortality in the hosts following stress conditions (Overstreet, 1973) and hence the need for further studies on the nature of their association have been stressed.

The present paper deals with the association of the peritrich *Zoothamnium rigidum* and the heterotrich *Stentor coeruleus* with the shrimp *Metapenaeus monoceros* and the tanaidacean *Apeudes chilensis* collected from the Cochin backwaters and reared in the laboratory. Another peritrich *Lagenophrys cochinensis* was observed on the tanaidacean. A microsporidean sporozoan was also found as endoparasite in these crustaceans.

MATERIALS AND METHODS

Samples of juvenile shrimps in drag nets and tanaidaceans using grabs were collected from Ramanthuruth area in Cochin and examined during premonsoon, monsoon and postmonsoon months. Live shrimps were reared in 40 litre plastic tanks containing filtered estuarine water and tanaidaceans were cultured in 10 litre plastic containers in which soft sediments enriched with decaying vegetation was provided as substratum. Shrimps were fed on artificial diet made of fish meal, tapioca flour and rice bran.

Isolated protozoans were fixed in Schaudin's, Bouin's, Zenker's and 4% formalin and stained with Heidenhain's haematoxylin and Mallory's triple connective tissue stain. Experiments were conducted in 10 litre glass aquaria using millipore filtered water.

Experimental infestation of *Z. rigidum* on *M. monoceros* was done, using 20 shrimps of 22 to 48 mm size (18 without and 2 with infestation) in one tank and another tank having 20 uninfested shrimps served as control. Shrimps were taken out on a plastic netting and washed with filtered water before introducing into test aquaria. Compounded diet mentioned above was used for feeding. The experiment lasting for four weeks was run in triplicate. Moults were also examined for associates.

To test the effect of short term stress produced by the increase in turbidity and depletion of oxygen on *Zoothamnium* infested tanaidacean culture, the aeration was stopped and the bottom sediment was stirred for 5 minutes and left as such for three hours and then the aeration was resumed. Dead individuals were collected and examined. The percentage of infestation in the population was recorded by sampling with cores. Oxygen level in the culture tank was estimated before and after stirring.

OBSERVATIONS

1. *Zoothamnium rigidum* (Precht)

Of the three species of ectocommensalic ciliates observed the heaviest infestation was that of *Z. rigidum*. The percentage frequencies of their natural occurrence in tanaidacean was the highest (29.8) during monsoon and followed during postmonsoon (4.54) and premonsoon months (1.96). Similarly in shrimps, rate of natural infestation was 18% in monsoon, 3.5% in postmonsoon and 1.6% in premonsoon samples.

The colonies of this peritrich was observed on all parts of the body including the eyes. Comparatively heavier infestation was noted at the anterior part of the hosts where upto 40 colonies/mm² of body surface were present. The myonemes of the colony were found interconnected so that the entire colony could contract or expand simultaneously.

The colonies measured upto 800 μ in length and the individual zooids measured 40–80 μ by 30–45 μ in size (Fig. 1). The sphinctor collar is double but its degree of development varies. The macronucleus is circular in diameter and often appeared twisted. The food vacuoles are often filled with some greenish substance, suspected to be of algal origin.

2. *Lagenophrys cochinensis* (Santhakumari and Nair)

They are found in large numbers on *Apseudes chilensis*. The heaviest infestation was 80–100 individuals/mm². About 15% of the tanaidacean were found infested during the monsoon. Rate of infestation varied between 4.5–6% in premonsoon and postmonsoon samples. The infestation was mainly concentrated on the posterior appendages though they were distributed on all parts of the body.

These are solitary loricate ciliates in which the peristomial disc lies at the tip of a stout neck which is the only part of the body to be extended through the mouth of the lorica. (Fig. 2). The lorica of the animal measures 60–90 μ in length, 50–65 μ in width

and is about 20–23 μ in height. The mouth of the lorica is 20–25 μ wide. Anterior and posterior lips are fairly developed. The posterior lip has a crescentic thickening resembling to a transversely folded membrane. The animal measures 50–65 μ in length and 20–28 μ in width. Macronucleus is oblong and lies closer to one side of the body. The central part of the body is occupied by a large number of food vacuoles. When the animal is disturbed it contracts the cilia, withdraws the neck and assumes a different position within the lorica.

3. *Stentor coeruleus* (Ehrenberg)

Stray specimens of *S. coeruleus* were found on various parts of the shrimp and tanaidacean during low saline period. This also occurs as free living in this area.

4. *Sporozoan*

An unidentified microsporidean resembling *Nosema nelsoni* Sprague was found to occur as endoparasite in *M. monoceros* and *A. chilensis*. The infection was detected in animals maintained in the laboratory and occasionally mass mortalities were observed in the host population. Heavily infected individuals come out of their burrows and crawl over the substratum till they die. Infected areas examined under the microscope showed clusters of this sporozoans. In some places they had replaced the muscle tissues. In a few shrimps the infection was detected only after maintaining them for a month in the laboratory. The infected tissue appears white in colour and the host becomes moribund as the infection advances.

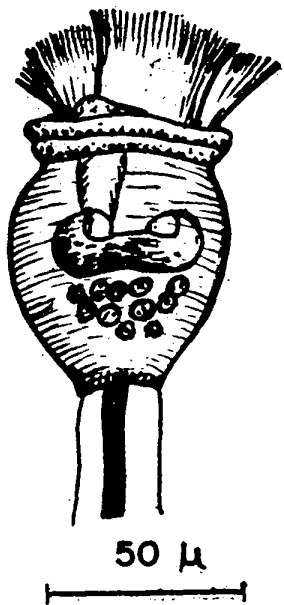


Fig. 1. *Zoothamnium rigidum*, a single zooid, showing the details of structure.

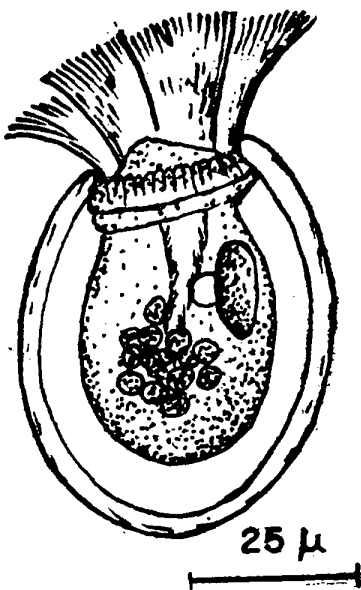


Fig. 2. *Lagenophrys cochinchensis*, showing the details of structure.

Experimental infestation of Z. rigidum

Experimental infestation of *Z. rigidum* carried out in *M. monoceros* showed that colonies of the ciliate get established on uninfested shrimp in a minimum of 3-4 days. Growth of the colonies and complete fouling of the surface of the shrimps takes place during the interecdysal period. On ecdysis the swarmer detach from the stalks to reinfest the fresh surface (Fig. 3). All the 54 shrimps used (18 per batch) in the experiment got infested by the contact of already infested individuals (Fig. 4). There was no infestation in the shrimps in control tanks.

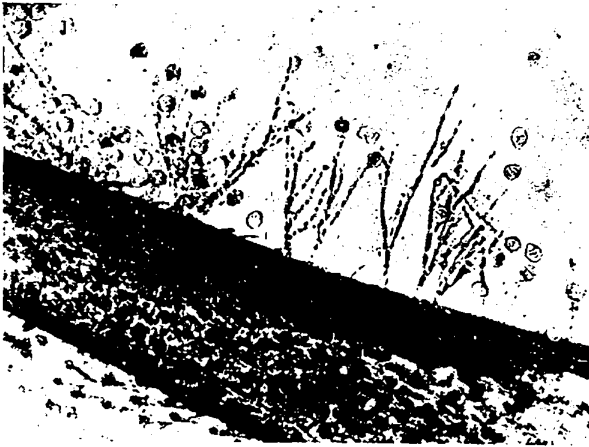


Fig. 3. Detached swarmer from the colony of *Zoothamnium* from the rostrum of the host (moult), *Metapenaeus monoceros*.

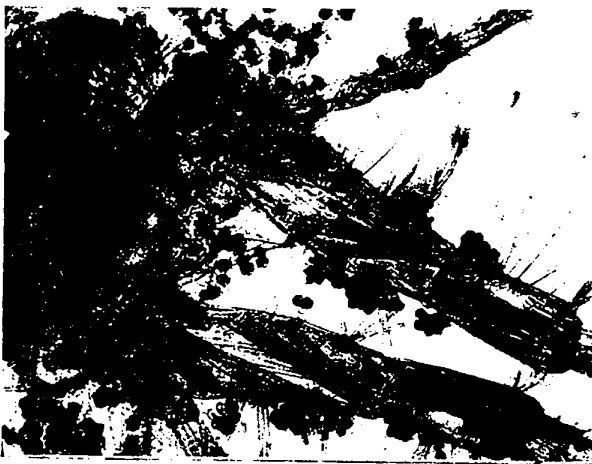


Fig. 4. *Z. rigidum* clusters attached on the anterior region of the host *Aapseudes chilensis*.

Effect of stress on Zoothamnium infested tanaidacean

Following the stress produced by the lowering of the oxygen level from 4.3 ml/l to less than 1 ml/l, large number of tanaidaceans began dying. Among the dead the percentage of infested was 81.81, 67.90 and 42.30 respectively.

As could be seen from the Table I a relatively high vulnerability to stress produced by high turbidity and low dissolved oxygen level is evident among the infested individuals. On an average 64% of the infested individuals were found dead on account of the stress produced by the lowering of oxygen level from 4.3 ml/l to less than 1 ml/l when the medium became highly turbid. Rate of mortality among the uninfested individuals came to an average of 6.13%.

DISCUSSION

Among the three species of ciliates found associated with shrimps and tanaidacean the most conspicuous was *Z. rigidum*, the heavy growth of which seems to deteriorate the chitinous exoskeleton of the hosts. It is interesting to note that the formation of free swimming swimmers of this sessile ciliate at the time of moulting, facilitates reinfestation on the new exoskeleton.

Relatively high rate of infestation, with more dense growth of larger colonies occurred on the reared individuals than on those collected from nature, which shows the probable conduciveness of stagnant water and over-crowding for infestation, as has been observed by Overstreet (1973). Similarly increased rate of infestation during monsoon is an indication to their preference to low salinity. The concentration of more ciliates on the anterior end of the hosts might be due to the need for taking advantage of the respiratory current of the host for feeding and respiration. This overcrowding seems to create obvious hindrance to the movements of hosts.

The majority of the *Zoothamnium* infested, tanaidaceans were found to be dead as a result of the artificially produced stress (Table I). Though seemingly harmless and as such non-parasitic in nature, *Z. rigidum* acts as a potentially harmful organism to its hosts. The deterioration of exoskeleton due to over crowding of this ciliate might serve as portals for other harmful organisms such as bacteria and fungi capable of producing serious shell diseases as has been observed by Rosen (1970) and Gopalan and Young (1975).

A large number of tanaidaceans that died as a result of artificial stress, were infested with *Z. rigidum*, and the rate of mortality varied from 42.3 to 81.81. The uninfested ones recorded a mortality between 2.9 and 10.57. This shows a possible predilection to

Table I. Effect of stress on tanaidacean infested by *Zoothamnium rigidum*.

Experiment	Total No.	% infested	No. infested	Total dead	%infested among dead	%dead among uninfested
No. I.	1150	12.43	144	176	81.81	2.89
No. II.	850	9.52	81	119	67.90	4.94
No. III.	357	7.28	26	61	42.30	10.57
Total	2357	—	251	356	—	—
Average	785.66	9.966	83.66	118.66	64.00	6.13

mortality of hosts as has been pointed out by Overstreet (1973). The over crowding of the ciliate in the anterior region of the hosts might be depleting oxygen from the already impoverished medium. Johnson (1972) has attributed instances of mortality of pond reared shrimp to infestation of *Zoothamnium*. However, Santhakumari and Nair (in press) have observed no ill-effects in the host *Sphaeroma* heavily infested with *Z. rigidum*. The profuse growth of these colonial ciliate might physically hamper the free movement of the hosts, particularly the young ones and make them more vulnerable to predators. Being a solitary form *L. cochinesis* does not seem to interfere with the locomotion of the host even though their encrustation produce scars on the exoskeleton. This species has been described as an ectocommensal on *Gammarus* (Fenchel, 1965). However, Couch (1966) reported that the infestation of *Lagenophrys* sp. causes harmful effects to crabs. The association of *S. coeruleus* seems to be purely casual, and is unlikely to produce any harmful effect in the host.

The microsporidean *Nosema* sp. is a typically parasitic form and has been reported in blue crabs and shrimps (Sprague, 1965 and Overstreet, 1973). The shrimps and tanaidaceans infected with the microsporideans, were obtained from the same locality but large scale infection and mortalities were noted only in tanaidacean cultures. Since shrimps are known to feed on tanaidaceans it is likely that the transmission of the parasite takes place by oral ingestion as in other sporozoans. However, this has to be confirmed by future studies.

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