PATHOLOGICAL CHANGES CAUSED BY PARASITES IN CHICOREUS RAMOSUS L.

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ABSTRACT
Sporocysts of trematodes and nematodes infested the foot, mantle, digestive gland (hepatopancreas) and gonad of the marine gastropod Chicoreus ramous. The digestive gland was the preferred site of infection. Visual examination of the foot revealed tissue reactions in the form of transparent blisters, mainly on the dorsal lateral sides due to infection by the parasitic helminth Echinococophalus sp. (Family Gnathostomidae). Numerous cysts of trematode parasites caused extensive damage to the digestive gland, but slight destruction of the gonad. Infected animals survived even though the digestive gland had been severely obliterated. Histopathological examination of the digestive gland revealed multifocal epithelial necrosis, autolysis and alteration in the hepatopancreatic tubules and mechanical destruction of cells. Infestation was more frequent in adults than younger ones. Most maics were subjected to chronic infection. The infected animals generally had a heavily damaged or bored shell.

INTRODUCTION
The role of marine snails as host of zooparasites has developed much interest in recent years because of far reaching implications. The host parastae relationship between the snails and the parasites have shown that many larval forms of the parasites, especially the cestodes, developed in molluscs and later in adults of various economically important species of fishes. Further the marine snails also serve as intermediate hosts to many other groups of parasites like protozoans, helminths and arthropods which act as carriers of pathogenic bacteria, viruses and other micro organisms of medical and veterinary importance (Cheng, 1967). The pervasive and severe effects of parasites on the population density of marine snails should not be underestimated since this type of association invariably leads to sterilization of the host, inhibits its seasonal migration and ultimately leads to its early death (Siddemum, 1970). Over and above, some marine snails have been reported to harbour larval stages of avian schistosomes which can invade the human skin causing dermatitis (Fenner, 1950; Leigh, 1952).

The predatory marine snail Chicoreus ramous L., 1758 of the family Muricidae was found to serve as intermediate host in many trematodes, nematodes, protozoans and other common inhabitants of the mantle of the host. However, knowledge about parasites and diseases of marine invertebrates, and pathogenicity of the parasites infecting C. ramous are inadequately characterised and probably have not been recognised. This family of gastropods has recently gained much economical, and also basic biological interest. The meat of the snails has good export value, and is also consumed by the rural population of India. Therefore, efforts were made to investigate the groups of parasites chiefly infecting these snails. This paper mainly concerns parasites infecting C. ramous and the alterations in the structure, size of chronic infection and the shell abnormalities caused as a result of the parasites.

MATERIAL AND METHODS
C. ramous were collected from Cuddalore and Nagapattinam waters on the southeastern coast of India. They were brought to the laboratory and maintained in well-aerated sea water. Later the animals were opened up and the tissues and muscles were examined under a binocular dissecting microscope.
The tissues found to be infected or parasitized were dissected out, and specimens were examined. The snails chosen for sacrifice were dipped into Bouin’s solution for 10 - 10 seconds to identify presence of sporocytes. Free living stages of the parasites were obtained from the media. The site of parasitic infestations and the larval stages of the parasites were photographed either alive or in the preserved state. For histopathological observation, the infected parts were fixed in Bouin’s fluid, embedded in paraffin wax (melting point 58°C), sectioned (4-5μm) and stained with haematoxylin and eosin. The parasites isolated were preserved by expanding them with a few drops of rectified spirit over the surface of the bodies, stained and mounted on a glass slide. The observations chiefly concerned the digestive gland, gonad, foot and mantle region of C. ramossus.

RESULTS

Generally, parasitics in C. ramossus were noted in pre-adults and adults rather than in younger ones. Males were found to be more frequently infected than females. Snails were infected by a number of common inhabitants like crustaceans and protozoans, but marked parasitism was caused by trematodes and nematodes in the foot, mantle, digestive gland and gonad. However, the most extensive damage and severe infestations were encountered in the digestive gland (hepatopancreas) region. This part, followed by the foot and mantle, was the preferred site of infestation by trematode parasites.

Foot

The foot portion of C. ramossus was infested with encysted larval forms of the nematode Echinostomoides sp. Many encysted larvae were identified in the head & foot region in the form of swellings or blisters aggregated in groups. They appeared as transparent white spots when located near the surface whereas those located deeper in the foot tissue appeared as dark spots. The blisters were more common in the dorsal-lateral sides of the foot and base on the ventral sides.

Digestive gland

The digestive gland complex was severely infested with numerous cysts of parasites. Sacrifice of several specimens revealed that the digestive gland was the primary site for encysted parasites. Tissue smears of the cysts showed emergence of metacercariae, a larval form of the trematodes inside the cyst. The cysts were orange or pale brown in colour, and embedded in various parts of the digestive gland. Encysted larvae appeared to be surrounded by a thin layer of connective tissue (capsule), perhaps of host origin. The occurrence of these encysted larval forms resulted in severe morphological changes of the digestive gland in the vicinity of the cyst. However, the animals were found to survive even though the digestive complex was severely obliterated.

Gonad

As a result of infestation by trematode cysts in the digestive complex, slight invasion and eventual destruction of the germinal epithelium were observed to have taken place in the gonad. The normal gonad development was slightly inhibited, and the gonad appeared emaciated. However, in most cases, no stress was found to occur in the gonad region though the digestive complex was infected.

Mantle

The mantle layer was mostly found to harbor a number of parasitic copepods and amphipods, but these parasites did not cause any pronounced affect like thromes that occurred on the digestive gland. However, slight scarring in the ventral side of the mantle was noted.

PATHOLOGICAL CHANGES

Histopathological examinations of the infected tissue sections (4-5μm) revealed the following changes in tissue morphology.

Foot

Cross sections of the infected foot part of C. ramossus revealed a generalized pattern of focal necrosis of the lining of epithelium. Severe ramifications and by-
six of the stromal tissue was observed. Lining epithelium became atrophied close to the area of blesters.

Digestive gland
Severe changes involving mechanical destruction of cells coupled with autolysis and lysis induced by the encysted parasitic larvae were noted. There were also marked pathological changes in the digestive tubules and the tubules which appeared to be completely or partly denuded, accompanied by marked infiltration of necrotic leukocytes. The damage resulted in alterations in the digestive epithelial cells, shape, and structural integrity. Random focal necrosis was noted near the periphery of encysted parasites. Dislocation of the surrounding hepatopancreatic tubules was caused by this infection.

Gonad
Changes in the gonad region were due to atrophy. The section showed slight histodysty of the glandular epithelium resulting in emaciation of the gonad.

Mantle
There was no marked pathological change but there was a slight pattern of ulcers in one or two places of infected area.

SHELL ABNORMALITIES
Massive destruction was found in heavily fouled shells. Most of the infected snails possessed a porous and heavily or moderately damaged shell, depending upon the degree of fouling. The shell was invariably honey combed causing excessive inner shell deposition and cracking. Small pores on the exterior of the shell were presumed to have occurred due to penetration of boring sponges, mostly of the genus Clione. Polycheates occurred in large numbers causing such shell abnormality. Outstanding in this respect were Polydora sp. which formed 'Mud blisters' and infiltrated into the shell. These worms were found to be the most dominant destructive agents of the Chitonus shell. Many shell boring bivalves of the genus Lithophaga were found to burrow inside the shell and to be scaled off inside the shell layer. These in course of time are presumed to bore through the inner shell. The incidence of boring by sponges was high in the spire region and the thickest part of the shells. The polycheate worms entered the shell through any opening made on the surface of the shell, and eventually burrowed into the shell. However, the exact type of association of these associated organisms could not be traced. Pores and destruction caused by these animals may render C. ramosus vulnerable to various other predators and microorganisms.

DISCUSSION
Though no parasites have been reported from the shell of molluscs (Cheng, 1967), certain shell boring bivalves that are capable of boring through the shell may pave way for the entry of parasites, thereby increasing the infection by parasites on the host. Among the internal groups of parasites in Chitonus ramosus the trematodes comprise the largest group followed by nematodes. Infection caused by the nematode Eubacchecephalans sp. in the form of transparent blisters revealed eroding of the epithelial layer and necrosis of tissues near the site of infection. This may decrease the efficiency of the foot as a rigid holdfast organ. The infected foot appeared to be shrunkken when removed from the animal. This response to invasion by larval helminths in snails has been reported to cause injury evacuation in gastropods (Sparks, 1972). Over all, human consumption of a diseased foot may lead to serious maladies, if the snails are not properly cooked.

Infestation caused by trematode parasites on the digestive gland and the adjacent gonad has been identified to cause dislocation of digestive tubules, autolysis, necrosis and eventual destruction of hepatopancreatic and the adjacent gonadal tissue. The phenomenon of autolysis is presumed to have taken place due to the stress caused to the small during cracking of the shell, or due to the preservation. It has been reported that trematode invasion and infestation of the digestive gland leads to degeneration of the gland, increasing mortality of an animal (Kime, 1980). However in this case, C. ramosus
seems to have survived even though the digestive gland was severely obliterated. Trematode invasion of the digestive complex caused some parasitic influence in the gonad region since the location of the gonad was adjacent to the digestive gland. Infection and parasitism have been reported to result in spontaneous reduction of the reproduction potential, like gonad and penis reduction, cessation of sperm and egg production since most of the nourishment of the digestive gland is being absorbed by the parasites. (Pan, 1965; McClelland and Bourns, 1969; Loker, 1979; Lysaght, 1941; Berry, 1962; James, 1964). Severe and frequent infestations by parasites of the adult males are attributed to the fact that they consume large quantities of food to satisfy the demands of the invading parasites. Convergence of the inflammatory cells in some regions of infection indicated that immunity developed by the cells against the infecting parasites.

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