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Histopathalogy of *Procamallanous lonis* (Yamaguti ,1935) from economically important fin fish within Batticaloa Lagoon.

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Abstract

Procamallanus lonis (Family Camallaidae) infecting Siganus sp. at higher prevalences was found to cause growth retardation. This study, therefore, focused principally on the histopathology caused by the above parasite on a population of Siganus from the Batticaloa Lagoon. The inflammatory response of the host provides sufficient clues to the pathogenicity of this parasite on economically important host; these are tissue invading parasites are more pathogenic rather than merely being intestine inhabiting parasites. The infections of Procamallanus lonis in Siganus sp., Glossogobious giuris, Tachysurus sp. and Ompak bimaculatus were studied to assess their pathological effect.

Infected tissue samples, notably from the intestine, liver and kidney were fixed in 10% formalin and processed for wax embedding. All tissues were cassetted, labeled and processed for histological sectioning. Five micrometre thick sections were cut and stained with haematoxlin and eosin. In the intestine infections elicited an inflammatory response leading to a light epitheloid encapsulation of the parasite. The encysted parasite causing pressure changes to the surrounding tissue. Heavy nematode infections were accompanied by hypertrophy of the tissue and the formation of a hyper plastic tissue reaction. Such marked tissue reactions may alter the absorptive nature of the intestine and presumably, the health condition of the host.

Key words: *Procamallanus lonis, Ompak bimaculatus, Siganus sp,* Batticaloa Lagoon, histopathology

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1. Introduction

The most important factor for selection of the host and a site of infecting by a parasite depends mainly on the nature of the defensive response elicited by the host against the parasite. The effect of the parasite and the effects of the host response results on host's survival are the most important factors for fish culture rather than the effects on the parasite.

The host response to parasites can be acute or chronic inflammatory response or a delayed or cell mediated response, which may provide sufficient clues to the pathogenicity of the infecting species and its impact on the host.

An inflammatory response leading to encapsulation of the parasite is common host reaction to the presence of nematode larvae [1]. The description of histopathology of the encysting larva of the nematode *Pseudoterranova decipiens* exhibited the same pattern [2]. It is not surprising that tissue invading parasites generally tend to be more pathogenic than those inhabiting the intestinal lumen.

Nematode larvae inflict direct damage to their hosts during the process of migration before they encysted within the space created within the invaded organ [3]. The pathogenicity of encysting larvae depends on the tissue in which they are present, the path of migration, the degree of penetration, the size of the encysted larvae and the parasites species in question. While many authors have detailed the degenerative changes in host tissues surrounding nematode infections tissue such as pressure atrophy, ascites and visceral adhesions and in severe cases, abdominal distension [4-7].

In this study the histopathological changes inflicted by nematode larvae within the intestines of siganids and other fish species were investigated to determine their potential impact on populations of these commercially important fish in Batticaloa Lagoon.
The main aim of this study, therefore, was to obtain a clearer understanding of the histopathological effect of *Procamallanus lonis* on fish hosts within Batticaloa Lagoon.

2. Materials and methods

Sampling procedure:-

Live specimens of *Siganus* sp, *Tachysurus* sp, *Glossogobious giuris* and *Ompak bimaculatus* were obtained from two locations within Batticaloa Lagoon. Fish were obtained from fisherman using cast nets in the sub-littoral zones of the lagoon. Sampling was carried out by two fishermen in each site who demonstrated extreme care in the capture of the fish to ensure mortalities were kept to a minimum.

Fish were transported to the laboratory at the South Eastern University in oxygenated polythene bags and maintained in the aquarium facility of the Biology Dept, in the same water in which the fish were caught until they were processed.

Parasitic survey

Samples of the gut, gills and liver were taken from a range of infected *Siganus* sp (n = 475), *Tachysurus* spp (n = 1152), *Glossogobious giuris* (n = 404) and *Ompok bimaculatus* (n=23) fixed in 10% buffered formalin, refrigerated for 24 hours to minimize tissue degeneration during the process of fixative penetration and then prepared for histological examination. All the tissues were trimmed, cassetted, labelled and processed for wax embedding following standard procedures. The wax infiltrated tissues were then positioned in suitably sized moulds, blocked in molten wax and cooled rapidly on a freezing plate. The blocks were then trimmed to expose the tissue and then microtomed to give 4-5µm sections were stained with haematoxlin and eosin. The sections were floated on water bath maintained at 40°C and collected on pre-washed glass slides.

3. Results

Procamallanus lonis encysted larvae were found in the abdominal cavities, livers, mesentries and intestines of *Siganus lineatus*, *Siganus oramin* and *Glossogobious giuris*, within the intestines of *Tachysurus* sp., and the intestines of *Ompok maculates* (Figure 1). The larvae were lightly encysted surrounded by host fibrous tissue. The capsules were approximately spherical, enclosing the larvae. Histological examination of cut sections show several transverse sections through the body of the nematode larvae although it is impossible to say whether this represents multiple sections through a single worm curled up within the cyst or multiple worms occupying the same cyst (Figure 2). Although the nematodes are likely to be curled within the cyst the precise number could only be determined by reconstruction of the cyst using serial sections.

An initial response to nematode infection has been through the infiltration of macrophages, as part of the inflammatory response, which consequentially has lead to epitheloid encapsulation (Figure 3). The cellular response which led to the formation of capsule was not clearly seen in some places. Cysts containing larvae were also found attached to the mesentery. No cysts within the liver were found during post-mortem examination.

Figure 1: An encysted of *Procamallanus lonis* attached to the intestine of *Glossogobius giuris* and damage. Scale bar = 0.1mm

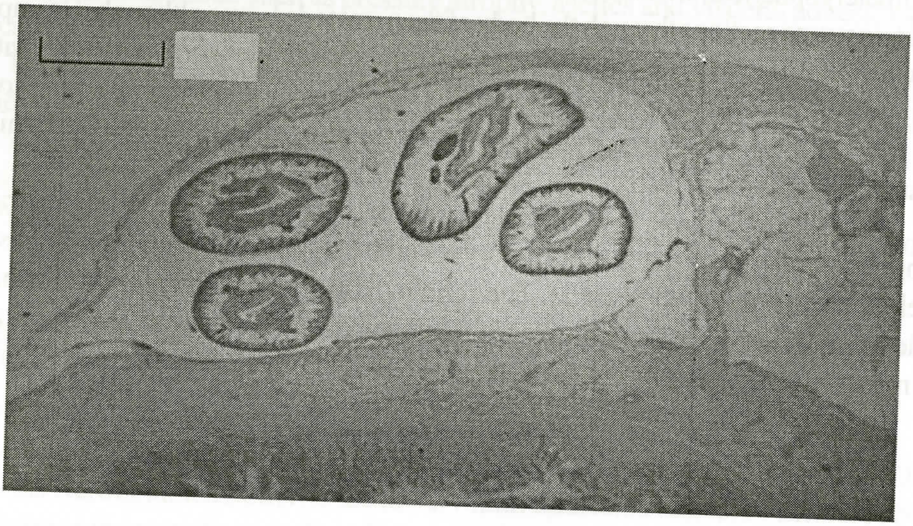

Figure 2: Transverse section through the intestine of a species of *Siganus* and lightly encysted nematodes. The cyst contents show four sections through the body of a nematode although this represents a single coiled individual or multiple worms within the same cyst would require confirmation by serial section reconstruction. Scale bar: 0.1mm

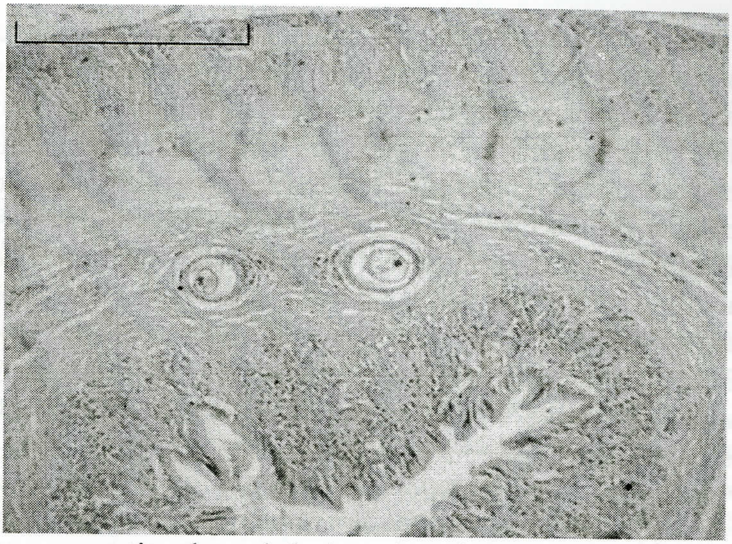

Figure 3: Transverse section through the intestine of *Siganus* spp. showing the presence of macrophages and the formation of capsules around two nematodes. Compression of the host tissue caused by the presence of the cysts can be seen. Scale bar: 0.1mm

4. Discussion

The effect of helminth infections within the liver of Atlantic cod (*Gadus morhua*) has been thoroughly reviewed by Margolis [9], where reductions in the size, weight and fat content of the liver, in the total weight of fish and in the condition factor of the fish were noted. The absence of data on the age, sex maturity, spawning condition and location and time of collection, however, makes interpretation of the field data difficult [9]. Paperna [10] also reported that parasitism reduces the condition factor of fish. The fish that were post-mortemed in the current study were in emaciated condition. The number of infected cysts were high in high distressed fish likewise it indicates the infection level through the physiological activities of the fish varying from active to lethargic.

The sequential involvement of inflammatory cells in the host response leading to the encystment of the metacercaria *Stephanochasmus baccatus* was eloquently demonstrated by Sommerville [11]. This has also been shown more recently by Nickol [12] and acanthocephalans which elicit an inflammatory reaction. Although this study was not able to study early infections, the production of a fibrous capsule to encapsulate invading nematodes was evident. The involvement of macrophages in the formation of epithelioid tissue and the laying down of fibrous cyst in this study, however, was clear.

There is no agreement as to the function of eosinophlic granular cells in fish. In higher animals, true eosinophils are often associated with parasitic infections. Eosinophilic granulocytes are reported in normal fish tissues such as within the epithelium (Roberts, Young and Milne [13]), the gills and the gut epithelium (Chaicharn and Bullock cited by Nicoll [14]: Ezeasor and Stokoe [15]: Ellis [16]. The presence of eosinophilic granular cells in the vicinity of inflamed infection sites, however, has been documented for some parasites. Lester and Daniels [17] found a large number of eosinophilic granulocytes in the inflammatory response of the white sucker (*Catostomus commersoni*), to the parasites *Actinobdella ineqsuiannulata* and *Lernaea cypyrinacea* L. (Shariff and Roberts [18]) and it seems likely that these are eosinophilic granular cells Noga [19]. In S*igunus* sp., eosinophilic granular cells were found at sites where *Procamallanus lonis* were found invading the host tissue (Figures 2 & 3). It was clear in the tissue reaction that eosinophilic granular cells appeared after invasion of the *Procamallanus lonis* larvae and production of the fibroblast capsule.

The absence of cysts in the liver might be suggested that this organ is not a usual site of infection or that cysts have been over looked if the cysts were embedded deep within the liver. Heavy infections in the intestine, however, lead to the destruction of the intestinal villi and necrotic and degenerative changes to the epithelium which Nickol [12] states can adversely affect the absorptive efficiency of the fish intestine. This in turn may deleteriously impact on the general health and growth of the host.

5. Conclusion

This study provides information on the host reaction and pathology to infection by the camallanid nematode *Procamallanus lonis* in a range of fish hosts found in Batticaloa Lagoon namely several species of siganid and *Tachysurus, Glossogobios giuris* and *Ompok maculates.* There are a limited number of studies on *Procamallanus lonis* and the pathology it inflicts on its host and further studies are needed to determine to what extent this parasite impacts on the swimming, orientation, growth and sexual maturation performance of the hosts it infects.

Similarly, a comprehensive study is required to determine the infection routes and the hosts involved.

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