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Screening of Sanguinicolid Trematodes on Cultured *Lates calcarifer* Fingerlings in Peninsular Malaysia

C.K. KHOO[1], B.C. KUA[2], A. MOHD-ADNAN[1] and A.M. ABDUL MUNIR[1*]

[1]School of Biosciences and Biotechnology
Faculty of Science and Technology
Universiti Kebangsaan Malaysia
43600 Bangi, Selangor, Malaysia
[2]National Fish Health Research Centre (NaFisH)
Fisheries Research Institute
11960 Batu Maung, Penang, Malaysia

Abstract

Sanguinicoliasis is the aetiology for blood fluke infections both in marine and freshwater fishes, and it presents an ongoing threat to the aquaculture industry. Consequently, disease surveillance is an essential and important procedure for evaluation of fish health in cultured fish. In this study, we report the prevalence of sanguinicoliasis on cultured Asian sea bass (*Lates calcarifer*) fingerlings. Sanguinicolid infection was detected from fresh gill biopsy on 10.0 to 18.5 cm total length cultured *L. calcalifer* fingerlings from three localities in Peninsular Malaysia by the presence of the miracidia and embryonated eggs embedded in the gills. The prevalence of infection of sanguinicoliasis were 20.0%, 11.1% and 6.7% for fingerlings from Kedah, Terengganu and Johor, respectively. Further study on the life cycle of this parasite, particularly the identification of the intermediate hosts, is required to be able to work toward host elimination and subsequent eradication of this disease. These results also serve as a baseline data for infection prevalence of sanguinicoliasis in *L. calcarifer* fingerling stages.

* Corresponding author. Tel.: +60 03 8921 5696
E-mail address: munir@pkrisc.cc.ukm.my

Short Communication

Mariculture of finfish is one of the fastest growing sectors in Asian aquaculture. Demand for high value fish species such as groupers, snapper, Asian sea bass are huge and provide high revenues. Aquaculture of the Asian sea bass (*Lates calcarifer*, Centropomidae) in South East Asia dominates the aquaculture industry and comprises 90% of the region's total aquaculture production (Hanafi et al. 1995).

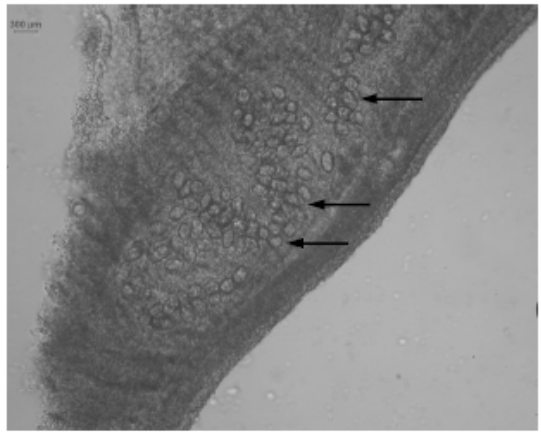
Diseases are one of the major risk factors in commercial aquaculture with millions of dollars lost annually (Shariff 1998). Like other farming systems, *L. calcarifer* aquaculture can be plagued with disease problems and numerous investigations on pathogens of *L. calcarifer* have been carried out (Chonchouenchob et al. 1987; Humphrey and Langdon 1987; Leong and Colorni 2002). One of the diseases threatening *L. calcarifer* production is the infection by endoparasitic sanguinicolid groups like *Cuoricola lates* (Herbert et al. 1994). Sanguinicolid or blood fluke is a contagious pathogen that infects the vascular system of both marine and freshwater fish (Bullard and Overstreet 2002). Fish may be carriers of sanguinicolid with no clinical signs, thus, establishing covert infections in fish with attendance economic consequences to aquaculture. This is especially true given that sanguinicoliasis can cause sudden mortality among cultured marine fishes (Ogawa and Fukudome 1994; Padrós et al. 2001). Therefore, fish disease surveillance is essential to the production of healthy fish and important to the evaluation of stock health in cultured fish. The aim of this study is to report the prevalence of the sanguinicoliasis on cultured *L. calcarifer* fingerlings from three localities within Peninsular Malaysia.

The Asian sea bass fingerlings (n = 202, total length: 10.0 – 18.5cm and weight: 13.0-54.0g) were collected and screened from geographically widespread localities comprising the northern locality (Tanjung Dawai, Kedah), eastern locality (Setiu, Terengganu) and southern locality (Tanjung Kupang, Johor) of Peninsular Malaysia. Fresh gill sections measuring 0.5 cm were examined by wet mount microscopic examination under a compound microscope (Leica DM5000 B). Signs of clinical disease were assessed and total numbers of sanguinicolid infected fish were recorded (Table 1). Among the three localities examined, Tanjung Dawai fingerlings showed the highest prevalence (20%) of sanguinicolid infections. Those from Setiu and Tanjung Kupang were reported to have 11.1 and 6.7% sanguinicoliasis prevalence, respectively.

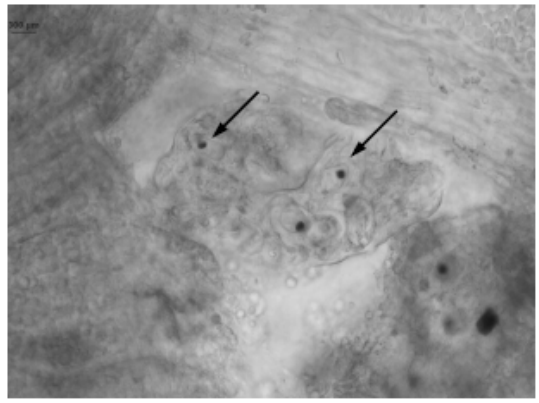
Table 1. The prevalence of sanguinicolid infection detected in Asian sea bass fingerlings (*L. calcarifer*) sampled from three different geographic localities within Peninsular Malaysia

Locality	Type of culture	Date screened	Total Length (cm)	Standard Length (cm)	Weight (g)	Total number of fish screened	Number of blood fluke	Prevalence of infection (%)
Tanjung Dawai, Kedah	Brackishwater earthen pond	30/03/05	10.0-12.0	8.5-10.0	13.0-20.0	25	5	20
Setiu, Terengganu	Pen	20/04/05	10.8-18.5	9.0-13.5	17.0-54.0	117	13	11.1
Tanjung Kupang, Johor	Brackishwater earthen pond	05/05/05	11.5-15.0	9.5-12.0	20.0-39.0	60	4	6.7

Sanguinicoliasis infections were revealed through the presence of both immature and mature miracidial stages as well the embryonated eggs embedded in the gill endothelium (Fig. 1a). The eggs were found to be round to oval shape and usually seen clumped. Clusters of eggs were mainly observed dispersed along the trailing edge of the gill lamellae. The developed miracidia were found actively moving around within the egg shell (Fig. 1b). From these observations, the miracidium appeared to penetrate through lamellar epithelium and subsequently eases through the gill epithelium swimming freely to the outside within seconds. The migration of the miracidium generated small empty breaks on the gill itself.



(a)



(b)

Figure 1. Different stages of sanguinicolid observed in Asian sea bass fingerlings (*L. calcarifer*) from aquaculture sites in Malaysia. (a) Mature and immature eggs of sanguinicolid observed on the fresh gill wet mounts (arrow), 10X; (b) Cluster of motile miracidia within individual egg shell (arrow), 20X

The presence of sanguinicolid in the three localities evaluated may be a reflection of the environmental state of each culture site. The brackishwater earthen impoundment culture system utilized in these localities offers both a concentration of fish and access to the intermediate hosts. Feed and waste products generated from these confined systems promote the propagation of the intermediate hosts. The abundance of mangrove swamp that became the habitat of all kinds of polycheate, bivalve and snails which could act as intermediate hosts for this blood fluke sited vicinity within the culture site might have promoted this endemic. These conditions provide a bountiful source of intermediate host for the parasite to propagate. For these reasons, these parasites managed to maintain their transmission successfully and survive. The result from this study indicated that reservoirs of sanguinicolids intermediate host were established within the localities screened.

The culture of many aquatic species often faces major setbacks due to disease infestation. This has greatly impeded both the economic and social development of many countries (Bondad-Reantaso et al. 2005). Survival of pathogens depends on, among others, host susceptibility and environmental factors influencing reproduction, growth and spread of the pathogen (Bakke and Harris 1998). In this study, the prevalence of infection of sanguinicoliasis were 20.0, 11.1 and 6.7% among 10.0-18.5 cm total length Asian sea bass fingerlings from Kedah, Terengganu and Johor, respectively. It is most probable that the intensification in the *L. calcarifer* aquaculture with the large-scale international and local movement of fertilized eggs, fingerlings or juveniles, as well as the rapid expansion and concentration of fish farms have facilitated the outbreak of these parasite infections within these localities. The data presented here contribute towards the understanding of the linkage of sanguinicolid trematodes fauna found in several aquaculture sites within Peninsular Malaysia. Also, this study results serve as the baseline report for the sanguinicoliasis in *L. calcarifer* fingerlings with total lengths ranging from 10.0 to 18.5 cm.

Sanguinicolid infections have been reported to cause either serious overall mortalities (Iqbal and Sommerville 1986) or a retarded growth performance of the infected fish (Evans 1974; Herbert et al. 1995). In this study the sanguinicolid infections observed were apparently mild and insufficient to cause lethal pathological effects. We make this judgement in that the infected individuals showed no overt disease, where in a heavy infection host blood loss from ruptured gill filament as a consequence of miracidial escape leads to mortality (Crespo et al. 1994). Impairment of the gill function may also be induced by embedded eggs due to physical

blockage of the blood flow subsequently causing mortality (Padrós et al. 2001). However either retarded growth at mild levels or mortality from heavy infections caused economic and productivity losses to farmers.

Prophylactic chemical treatments helped to reduce sanguinicoliasis and the parasite population but did not eliminate them totally (Bullard and Overstreet 2002). This is due to the sanguinicolid's heteroxenous life cycle that involves alternating sexual and asexual cycles in succeeding different types of hosts. Adult flukes were protected from prophylactic chemical as they resided in internal organs like mesenteric blood vessels, kidney, pericardial vessels and eye of the fish (Herbert et al. 1995). Only the free swimming stages of the miracidia and the cercaria are vulnerable to treatment. However, once the miracidium or cercarium penetrates into a suitable host, it becomes safe from the chemical treatment. Therefore, elucidating this parasite's life cycle, as well as, the identification of the intermediate host are essential for disease control. By maintaining good management that involves identification and elimination of susceptible intermediate hosts from the vicinity of fishes will subsequently eradicate this disease by breaking its complex life cycle (Bullard and Overstreet 2002).

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References

Bakke, T.A. and P.D. Harris. 1998. Diseases and parasites in wild Atlantic salmon (*Salmo salar*) populations. Canadian Journal of Fisheries and Aquatic Sciences 55:247–266.

Bondad-Reantaso, M.G., R.P. Subasinghe, K. Ogawa, S. Chinabut, R. Adlard, Z. Tan and M. Shariff. 2005. Disease and health management in Asian aquaculture. Veterinary Parasitology 132:249-272.

Bullard, S.A and R.M. Overstreet. 2002. Potential pathological effects of blood flukes (Digenea: Sanguinicolidae) on pen-reared marine fishes. Proceedings of the 53rd Gulf and Caribbean Fisheries Institute, pp 10-25.

Chonchouenchob, P., S. Sumpawapol and A. Mearoh. 1987. Diseases of cage cultured sea bass (*Lates calcarifer*) in south western Thailand. ACIAR Proceedings No. 20:94-197.

Crespo, S., A. Grau and F. Padrós. 1994. The intensive culture of 0-group amberjack in the western Mediterranean is compromised by disease problems. Aquaculture International 2:262-265.

Evans, W.A. 1974. Growth, mortality and hematology of cutthroat trout experimentally infected with the blood fluke *Sanguinicola klamathensis*. Journal of Wildlife Diseases 10:341-346.

Hanafi, H.H., M.A. Arshard and S. Yahaya. 1995. *Annex II Country Reports – Malaysia*. In: Regional Study and Workshop on the Enviromental Assessment and Management of Aquaculture Development (TCP/RAS/2253). NACA Enviroment and Aquaculture Development series No. 1. Network of Aquaculture Centres in Asia-Pasific, Bangkok, Thailand.

Herbert, B.W., F.M. Shaharom-Harrison and R.M. Overstreet. 1994. Description of a new blood fluke, *Cruoricola lates* n. g., n. sp. (Digenea: Sanguinicolidae) from sea bass *Lates calcarifer* (Bloch, 1790) (Centropomidae). Systematic Parasitology 29:51-60.

Herbert, B.W., F.M. Shaharom and I.G. Anderson. 1995. Histopathology of cultured sea bass (*Lates calcarifer*) (Centropomidae) infected with *Cruoricola lates* (Trematoda: Sanguinicolidae) from Pulau Ketam, Malaysia. International Journal for Parasitology 25:3-13.

Humphrey, J.D. and J.S. Langdon. 1987. Pathological anatomy and diseases of barramundi (*Lates calcarifer*). ACIAR Proceedings No. 20:198-203.

Iqbal, N.A.M. and C. Sommerville. 1986. Effects of *Sanguinicola inermis* Plehn, 1905 (Digenea: Sanguinicolidae) infection on growth performance and mortality in carp, *Cyprinus carpio* L. Aquaculture and Fisheries Management 17:117–122.

Leong, T.S. and A. Colorni. 2002. *Infectious diseases of warmwater fish in marine and brackish waters.* In: Diseases and disorders of finfish in cage culture (ed. P.T.K. Woo, D.W. Bruno and L.H.S. Lim), pp. 193-230. CABI Publishing, London.

Ogawa, K. and M. Fukudome. 1994. Mass mortality caused by blood-fluke (Paradeontacylix) among amberjack (*Seriola dumerili*) imported to Japan. Fish Pathology 19: 265-269.

Padrós, F., C. Zarza and S. Crespo. 2001. Histopathology of cultured sea bream *Sparus aurata* infected with sanguinicolid trematodes. Diseases of Aquatic Organisms 44:47-52.

Shariff, M. 1998. Impact of diseases on aquaculture in the Asia–Pacific region as exemplified by epizootic ulcerative syndrome (EUS). Journal Applied Ichthyology 14:139–144.

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