UNIVERSITI PUTRA MALAYSIA

MORPHOLOGY OF A SANGUINICOLID BLOOD FLUKE, *SANGUINICOLA ARMATA*, IN GRASS CARP (*CTENOPHARYNGODON IDELLA C. V.*) FINGERLINGS AND ITS PREVALENCE

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FPSS 1994 2

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MASTER OF SCIENCE
UNIVERSITI PERTANIAN MALAYSIA
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By

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Thesis submitted in partial fulfilment of the requirement for the Degree of Master of Science in the Faculty of Fisheries and Marine Science, Universiti Pertanian Malaysia

November 1994

ACKNOWLEDGEMENTS

I am immensely grateful to my supervisors, Dr. Faizah Shaharom, Associate Professor Dr. Rehana Abdullah Sani and Professor Dr. Mohamed Shariff bin Mohamed Din for their valuable and continuing supervision and advice. It is my pleasure to extend a special token of appreciation to Dr. Faizah Shaharom, the chairman of the supervisory committee for her encouragement, support and untiring guidance throughout the project.

My sincere thanks also to Professor J. W. Lewis of Royal Holloway, University of London, Professor Jiang Yulin of Institute of Hydrobiology, People Republic of China, and Dr. B. W. Herbert of Department of Primary Industries, Australia for giving some of the literatures needed for review. Special thanks to Professor Y. L. Song and Dr. C. Y. Chien of National Taiwan University, Taiwan for their efforts to cite literature of sangunicolid blood flukes in Taiwan.

My appreciation to Professor Bjohn Berland of Institute of Zoology, University of Bergen, Norway and Marcella, fiancee of Dr. Paul Van Mele of Faculty of Veterinary Medicine, Universiti Pertanian Malaysia for translating some of the literatures in German.

I am also indebted to Dr. Nadzri Salim, Faculty of Veterinary Medicine and Dr. Japar Sidek Bujang, Faculty of Fisheries and Marine Science for their advice and guidance on the use of epidemiological terms and statistical analysis respectively.

I greatly appreciate the assistance of Mr. C. C. Lee of the Faculty of Veterinary Medicine, Universiti Pertanian Malaysia, Mr. M. Krishnasamy of Institute Medical Research, Kuala Lumpur and Miss Si Si Hla Bu of School of Biological Sciences, Universiti Sains Malaysia for providing me some of the stains and staining procedures.

Thanks also to Puan Aminah Jusoh and Mr O. K. Ho from the Faculty of Veterinary Medicine for their technical assistance in Scanning Electron Microscopy and photography and to Encik Kadir bin Alias, the artist of Faculty of Fisheries and Marine Science for his final touch on the composite drawing of the adult fluke.

I thank the International Development Research Centre of Canada (IDRC) for financing the theory and research components of the masters programme. My sincere gratitute go to the Department of Veterinary Service, Ministry of Agriculture, Malaysia for granting me the opportunity to undertake this study at Universiti Pertanian Malaysia.

I thank all my friends and colleagues at the Universiti Pertanian Malaysia who helped me in various ways during the project.

Finally, I thank my husband Ramon Teo, my son Eng Chuan and my daughter Wen-jing from the bottom of my heart, for their unselfish sacrifices, patience and reassurance which made this endeavour a success.

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LIST OF ABBREVIATIONS

B - Bristles

BL - Body Length

BS - Blunt Spines

BW - Body Width

c - Caeca

cm - centimeter

cp - Cirrus Pouch

CP - Circular Papillae

EL - Length of Oesophagus

EP - Excretory Pore

ev - Excretory Vesicle

fgp - Female Genital Pore

gp - Genital Pore

h - Hour

Km - Kilometer

mgp - Male Genital Pore

MMC - Melano-macrophage centre.

mo - Muscular Organ

MOL - Length of Muscular Organ

MOW - Width of Muscular Organ

ms - Marginal Spines

mt - metric tonne

o - Ovary

oe - Oesophagus

od - Oviduct

OL - Length of Ovary

oo - Ootype

OoD - Distance of Ootype

ov - oviduct

OW - Width of Ovary

P - Proboscis

pp - Protrusible Proboscis

SEM - Scanning Electron Microscopy

SL - Length of Spine

sv - Seminal Vesicle

SW - Width of Spine

t - Testis

TL - Length of Testis

TMO - Terminal Mouth Opening

TS - Tegumental Spines

um - Micrometer

v - Vitellaria

vd - Vitelline duct

Abstract of thesis submitted to the Senate of Universiti Pertanian Malaysia in partial fulfilment of the requirement for the degree of Master of Science.

MORPHOLOGY OF A SANGUINICOLID BLOOD FLUKE, *SANGUINICOLA ARMATA*, IN GRASS CARP (*CTENOPHARYNGODON IDELLA* C. V.) FINGERLINGS AND ITS PREVALENCE.

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November 1994

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Sanguinicola armata Plehn, 1905 is redescribed under light and Nomarski microscopy and SEM (Scanning Electron Microscopy). It is a lanceolate sanguinicolid with a single column of heavy lateral marginal spines; extensive vitellaria; long oesophagus and an X-shaped intestine with five-lobed caeca. There are 9-10 pairs of testes; median; post intestinal caeca and a single butterfly-shaped ovary. Male and female genital pores lie adjacent to each other, near to the posterior extremity. The uterus is short, post ovarian and contains only one egg.

SEM study revealed minute, blunt spines at the posterior part of the proboscis and tiny posteriorly directed marginal spines at the posterior end which were not visible under the ordinary light and Nomarski microscopy. This had added extra taxonomical features to *S. armata.*

Surface morphology of *S. armata* under SEM also showed many tegumental folds with ridges and crest, cilium-like structure and sensory papillae which may be related to their nutritional, sensory, excretory, secretory, osmoregulatory and immunological functions. Protrusion of the genital pores viewed under SEM helps to understand the fact that cross-fertilisation as being a rule in the trematodes' reproduction.

The prevalence rate of infection of *S. armata* was found to be high (66.7%) in the locally produced grass carp (*Ctenopharygodon idella*) fingerlings in the farm under the present study. This was attributed to the abundance of aquatic snails which act as intermediate hosts of the fluke. However, the intensity of infection was low due to the host (immunological), environmental (stocking density and water chemistry) and parasite (reproductive biology) factors.

Since the distribution of *S. armata* was highest (87.8%) in the bulbous arteriosus, it should be examined first for quick diagnosis of the fluke.

Abstrak tesis yang dikemukakan kepada Senat Universiti Pertanian Malaysia, sebagai memenuhi sebahagian daripada keperluan untuk mendapat Ijazah Master Sains.

MORFOLOGI FLUK DARAH SANGUINICOLID, *SANGUINICOLA ARMATA* DALAM ANAK IKAN KAP RUMPUT (*CTENOPHARYNGODON IDELLA* C. V.) DAN PREVALENS PARASIT.

Oleh

ONG BEE LEE

November 1994

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Fakulti: Perikanan dan Sains Samudera

Morfologi *Sanguinicola armata* Plehn, 1905 dihuraikan semula dengan bantuan kompaun mikroskop menggunakan lensa biasa dan Nomarski serta SEM (Elektron Mikroskop Pembiadas). Ia adalah fluk darah sanguinicolid yang berbentuk daun dengan satu deretan spina yang tebal di bahagian pinggiran ventral; mempunyai vitellaria yang meluas; esofagus yang panjang dan usus yang berbentuk silang dengan lima lobus seka. Ia juga mempunyai 9-10 pasang testis, terletak di median, di bahagian belekang kepada lobus seka dan mempunyai satu ovari yang berbentuk macam kupu-kupu. Liang-liang genital jantan dan betina adalah bersebelahan antara satu sama lain, di bahagian belakang badan. Rahimnya pendek, terletak di belakang ovari dan mengandungi sebiji telur sahaja.

Kajian SEM membolehkan spina halus dan tumpul, kelihatan di bahagian posterior probosis dan spina kecil menghadapi posterior di bahagian belakang fluk yang tidak dilihat dengan mikroskop kompaun lensa biasa dan Nomarski. Ini menambahkan ciri taxonomi kepada fluk darah *S. armata.*

Morfologi permukaan *S. armata* di bawah SEM juga menunjukkan lipatan tegumen dengan rabung dan krista, struktur semacam silium dan papilla deria yang boleh dikaitkan dengan fungsi pemakanan, deria, perkumuhan, rembesan, osmokawalaturan dan imunologi. Liang-liang genital yang tersembul dari permukaan tegumen fluk membantu kami memahami bahawa persenyawaan silang adalah fenomena kebiasaan fluk.

Prevalens *S. armata* pada anak ikan kap rumput tempatan adalah didapati tinggi (66.7%) dalam kajian ini. Ini boleh dikaitkan dengan kehadiran banyak siput air di kolam sebagai perumah perantaraan fluk. Walau bagaimanapun pengamatan jangkitan adalah rendah dan ini mungkin disebabkan oleh faktor-faktor perumah (kesan imunologi), persekitaran (kadar pelepasan dan kimia air) dan parasit (biologi pembiakan).

Oleh kerana taburan *S. armata* adalah tertinggi (87.8%) di bulbus arteriosus, organ ini patut dipemeriksa dahulu untuk membuat diagnosa segera fluk ini.

CHAPTER I

GENERAL INTRODUCTION

Background

Aquaculture plays an important role in contributing towards our protein food supply and will become increasingly important as a response to increasing demand for fish and declining fish catches from natural resources.

In Malaysia, though aquaculture is a relatively young industry, it has developed rapidly in recent years. The total aquaculture production was 80,000 mt valued at RM 207 million in 1992 (Malaysia, 1993). This was about a 50% increase compared to the 1990 production.

Topping the list of production was cockle (*Anadara granosa*) followed by freshwater fishes. The major species produced were tilapia (*Oreochromis niloticus*) 4,631 mt; Javanese carp (*Puntius gonionotus*) 2,505 mt; common carp (*Cyprinus carpio*) 1,703 mt; bighead carp (*Aristichthys nobilis*) 1,222 mt; grass carp (*Ctenopharyngodon idella*) 913 mt; walking catfish (*Clarias* spp.) 904 mt; marble goby (*Oxyleotrix marmoratus*) 115 mt and the latest addition, eel (*Anguilla japonica*) 1,572 mt (Malaysia,1993).

Rapid development and intensification of aquaculture practices together with the degradation of the aquatic environment has resulted in increased incidences of fish disease. Economic losses due to disease can be reduced through adequate fish health management. However, in order to apply proper health management and disease contol measures, identification of pathogens and a clear knowledge of their biology are essential and must not be neglected.

Parasites play a major role among the pathogens which causes fish diseases in Malaysian aquaculture. The parasitic diseases that have been reported were ichthyophthiriasis; chilodonellosis; lernaeosis; argulosis (Shariff 1980; 1984), gold rust (Shaharom et al., 1990) and myxosporiodiosis (Shariff, 1982).

Sanguinicoliasis caused by sangunicolid blood flukes have been reported from many parts of the world causing high mortalities in hatcheries and farms. Severe epizootics in cultured fish due to *Sanguinicola* have demonstrated the importance of this parasite. There is no treatment indicated and control of the disease is by interrupting the parasite's life-cycle.

Sanguinicolids are blood flukes which live and reproduce within the circulatory system of fish. They are the only digeneans in which the adults are of economic importance to fish culture (Smith, 1972; Bauer et al., 1973). Most adult digeneans live in organs which have a direct connection to the outside, allowing eggs to pass out with minimal or no effect on the host. As blood

flukes live and deposit eggs in the circulatory system, the eggs ultimately settle and lodge in tissues, often causing severe pathological responses in host tissues. Hence, host-parasite relationships involving blood flukes usually differ from those of the majority of digeneans which live and deposit eggs in their host's alimentary tract and elicit little response.

The importance of fish parasites is related to the importance of the fish that they may affect. Sanguinicolids are very important parasites in the culture of many fish species world-wide including cyprinids in Europe, Asia and USSR; salmonids on the west coast of North America and siluroids in Africa. However, very little is known about their presence in fish from the Southeast Asian region probably because their presence is often overlooked as they are very small and not easily differentiated from the host tissues. *Sanguinicola armata* was first reported in bighead carp and grass carp fingerlings imported into Malaysia from Taiwan (Anderson and Shaharom-Harrison, 1986). However no work has been conducted to identify and to examine the prevalence of this parasite in fingerlings produced from Malaysia.

Significance of Grass Carp

Grass carp, *Ctenopharyngodon idella* Cuvier and Valenciennes 1884, was the fish species chosen for the present study. It is the most cultivated species in the Southeast Asian region (Pillay, 1976) and it fetches a high price in the market (Shireman and Smith, 1983). It has been the subject of considerable

research in recent years because of its ability to consume aquatic vegetation (Shireman et al., 1977; Riley, 1978 and Meske, 1985). Grass carp is susceptible to diseases and according to Wu (1971), fish farmers in Taiwan had their profits reduced each year due to high mortalities in cultured grass carp. A sanguinicolid blood fluke, *Sanguinicola magnus* was identified by Hu et al. (1965) causing high mortalities of grass carp fingerlings in China. However, there is no report of blood flukes from Taiwan and other Southeast Asian countries besides the report by Anderson and Shaharom-Harrison (1986) in the imported carp fingerlings in Malaysia.

In Malaysia grass carp is one of the major Chinese carps introduced in the 1800's (Welcomme, 1981). The introduction was associated with the immigration of Southern Chinese who brought along the techniques of culture (Ang et al., 1989). This fish has since become the mainstay of the local freshwater aquaculture and can be induced to breed in captivity. They are cultured in earthen ponds, mostly by small scale operators using polyculture and integrated farming system. They contributed significantly to the total freshwater aquaculture production in Malaysia. Percentage contributions from the major Chinese carps remained above 70% in the early eighties accumulating to a peak of 90% in 1984. However, their dominance is gradually being overtaken by tilapias in the nineties (Ismail, 1994).

Objectives

Culture of fish provides an ideal opportunity for the transmission of parasites due to the density of potential host and their confinement in a restricted area. Blood flukes are of no exception. Although the normal levels of infection as experienced in natural habitats do not appear to detrimentally affect the host, high levels of infection can result in reduced production and mass mortalities in farms. Outbreak of sanguinicoliasis in fish propagating ponds resulting in adverse effects on growth and survival have been reported in many parts of the world (Iqbal and Sommerville, 1986). Therefore, blood flukes is a cause of concern for freshwater aquaculture as it has not been particularly identified in locally produced carp fingerlings. Hence, there is a clear need to investigate this blood fluke to determine its effects on carp culture in Malaysia and the objectives of this study are as follows:

(i) to study the prevalence and intensity of infection of this parasite in locally produced grass carp fingerlings,

(ii) to describe and to identify the blood fluke, and

(iii) to study its surface morphology by scanning electron microscopy.