EFFECTS OF PROBIOTICS ON THE GROWTH AND SURVIVAL OF BLACK TIGER SHRIMP POST LARVAE (*Penaeus monodon*) IN RECIRCULATION WATER SYSTEM

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THIS DISERTATION IS SUBMITTED IN PARTIAL FULLFILMENT OF THE COURSEWORK TO OBTAIN BACHELOR OF SCIENCE (HONORS).

AQUACULTURE PROGRAMME
SCHOOL OF SCIENCE AND TECHNOLOGY
UNIVERSITY MALAYSIA SABAH

2009

KESAN PROBIOTIK TERHADAP PERTUMBUHAN DAN KEMANDIRIAN PASCA LARVA UDANG HARIMAU (*Penaues monodon*) DALAM SISTEM AIR KITAR SEMULA

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TESIS INI DIKEMUKAKAN UNTUK MEMENUHI SEBAHAGIAN DARIPADA SYARAT MEMPEROLEHI IJAZAH SARJANA MUDA SAINS DENGAN KEPUJIAN

PROGRAM AKUAKULTUR
SEKOLAH SAINS DAN TEKNOLOGI
UNIVERSITI MALAYSIA SABAH

2009

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I declare that this dissertation is the result of my own independent work, except where otherwise stated.

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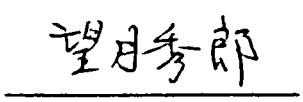

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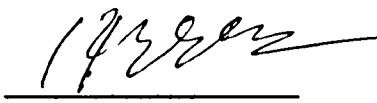

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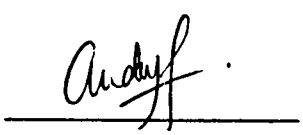

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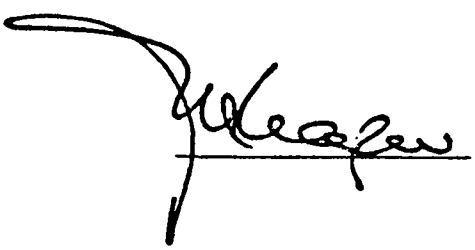

ACKNOWLEDGEMENTS

This dissertation has accumulated many debts of gratitude throughout its completion. Thus I would like to extend my heartiest appreciation to a great number of people who have contributed in my conquest of completing this dissertation. This dissertation would not have been successful without their full commitment. Firstly, I would like to thank my supervisor Dr. Annita Yong Seok Kian for her kindly who has been a great help support and guiding in my research for a great help in constructive advices on my work, valuable suggestions, continuous encouragement at various stages throughout the completion of dissertation also for her expert guidance and for her patience, without it this work could not have been accomplished. Not to forget Mr. Julian Rangsangan and his staff in the Microbiology Laboratory for their support. I am also greatly in debt for Dr. Hideo Mochizuki, Mr. Abentim Estim for their valuable advices, teachings and guidance. Lastly Higashimaru Co. Ltd. for preparing the feed.

ABSTRACT

Probiotics treatment on black tiger shrimp (*Penaues monodon*) treatments was carried out in 7-liter square aquarium and 240 tails shrimp post larvae (PL) were used in this trial. The trial was done in duplicate. In this study, treatment A was representing an additional of probiotics Bio-Aqua and nitrifying bacteria, treatment B was representing an additional of probiotics Bio-Block and nitrifying bacteria, treatment N representing an additional of nitrifying bacteria and treatment C as a control group without any additional of probiotics or nitrifying bacteria. A number of 30 tails of PL were randomly distributed into each of the four different treatment groups. Probiotics contain of *Bacillus* sp. was added to treatment A and B tanks. Survival rate and weight gain among three different treatment groups and control were determined at 18, 31 and 63 days after rearing. There was significance different ($P<0.05$) of survival rate and body weight gain of PL in probiotics supplementation groups. There was significance different ($P<0.05$) in water quality for ammonia, phosphate, nitrite and nitrate. Only nitrite test showed no significant difference ($P>0.05$) occurred between treatment groups A, B and N with control group. Overall, better water quality of ammonia, phosphate and nitrate was achieved in treatment A, B and N groups. After challenging PL with pathogen *Vibrio* sp., by immersion for 14 days, probiotics group B had 90% of survival rate. In addition the PL in probiotics treatment appeared healthy and active compared to treatment N and control.

ABSTRAK

Kajian terhadap udang harimau (*Penaues monodon*) telah dijalankan dengan menggunakan akuarium berkapasiti 7-L. Sebanyak 30 ekor larva telah dipilih secara rawak dan dimasukkan ke dalam setiap akuarium untuk empat kumpulan rawatan. Setiap kumpulan rawatan dijalankan dalam duplikat. Tangki rawatan A telah dimasukkan probiotik Bio-Aqua dan bakteria nitrifikasi. Tangki rawatan B pula dimasukkan probiotik Bio-Block dan bakteria nitrifikasi. Manakala tangki rawatan N hanya dimasukkan bakteria nitrifikasi dan tangki kawalan C adalah tanpa sebarang probiotik ataupun bakteria nitrifikasi. Tangki rawatan A dan B telah diberi probiotik yang mengandungi spesis *Bacillus* sp. Kadar hidup dan penambahan berat di antara kumpulan rawatan dan kawalan telah ditentukan pada hari ke 18, 31 dan 63 selepas rawatan. Terdapat perbezaan bererti ($P<0.05$) di antara kumpulan rawatan berbanding kumpulan kawalan bagi kadar hidup dan penambahan berat badan larva. Terdapat perbezaan bererti ($P<0.05$) mutu air bagi kepekatan ammonia, phosfat dan nitrat antara kumpulan rawatan. Hanya bacaan nitrit tidak menunjukkan perbezaan bererti ($P>0.05$) di antara kumpulan rawatan dan kawalan. Secara keseluruhan, mutu air bagi kumpulan rawatan kumpulan A, B dan N didapati lebih baik berbanding kumpulan kawalan. Bagi mewujudkan keadaan patogenik, *Vibrio* sp. telah di masukkan ke dalam tangki dengan menggunakan kaedah rendaman selama 14 hari. Kumpulan rawatan B telah menunjukkan kadar hidup sebanyak 90%. Tambahan lagi, udang dalam kumpulan probiotik telah menampakkan lebih sihat dan aktif berbanding kumpulan rawatan N dan juga kumpulan kawalan.

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LIST OF ABBREVIATIONS

°C	degree centigrade
%	percentage
ANOVA	Analysis of Variance
BW	Body Weight
cm	centimeter
DO	Dissolved Oxygen
FAO	Food and Agriculture Organization
g	gram
h	hour
ℓ	liter
BMRI	Borneo Marine Research Institute
PL	post larvae
ppm	part per million
ppt	part per thousand
RAS	Recirculation Aquaculture System
rpm	round per minute
sp	species
SPSS	Statistical Package for Social Science
TL	Total Length
UMS	Universiti Malaysia Sabah

CHAPTER 1

INTRODUCTION

1.1 Shrimp Farming in Aquaculture Industry

Shrimp farming has been one of the fastest growing aquaculture sectors in Asia and Latin America, and recently Africa. Global shrimp production as well as trading values and volumes have grown significantly in the past 20 years (Figure 1.1). With this rapid growth, there has been a concurrent and increasing demand for improved sustainability of shrimp aquaculture, social acceptability, and improved quality and safety of products produced by the sector (FAO, 2007).

Rapid expansion of shrimp farming has generated substantial income for many developing countries, as well as developed countries, but has been accompanied by rising concerns on environmental and social impacts of development especially on water pollution. Production of an aquatic animal in a large scale (commercial) may expose the shrimps to stressful condition, which tend to relate with disease outbreak and deterioration of environmental condition (Balcazar *et al.*, 2006a).

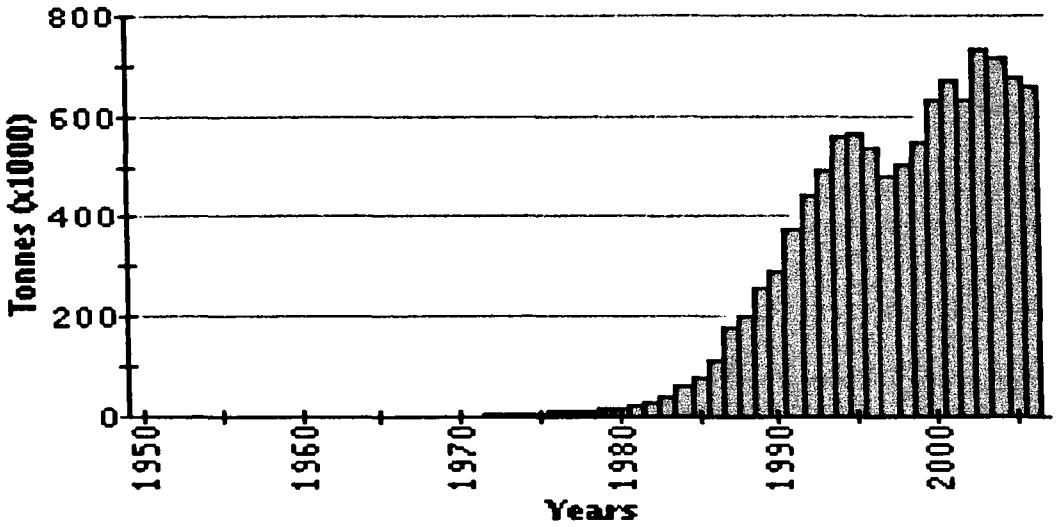

Figure 1.1: World production of *Penaeus monodon* in the world from year 1972-2006.

Source: FAO, 2007

Major environmental issues raised including the ecological consequences of conversion of natural ecosystems, particularly mangroves, for construction of shrimp ponds polluted the coastal waters due to pond effluents, biodiversity issues arising from collection of wild brood and seed, and social conflicts in some coastal areas. Aquaculture activities would intensively increase the occurrence of disease (Shariff *et al.*, 2001).

1.2 Black Tiger Shrimp, *Penaeus monodon* Industry Status

1.2.1 Current and Future Status of Black Tiger Shrimp Farming

In Malaysia, shrimp hatchery and nursery system are operating towards large scale production amount of seeds to be supplied locally and internationally. The developments of shrimp hatchery and nursery system are among the way to ensure the consistent supply of shrimp to support the shrimp industry. The FAO estimates that half of the world's seafood will be supplied by aquaculture within the next 10 years. Until now the production is considered low upon demand and overexploited of natural resources (FAO, 2007).

Photo 1.1: Black tiger shrimp (*Penaeus monodon*) in dark and white stripe above and blue reddish below; body with a six series of large segmented abdomen and carapace. Scale (1:2cm)

Shrimp farming play important role in economic activity in many countries especially in tropical area by offering poverty alleviation, employment, community development, reduction of stress towards natural resources over exploitation.

Even though black tiger shrimp, *Peneaus monodon* production is declined after infection some disease including; yellow-head baculovirus, luminous bacterium that affected worldwide, but still they are still the most widely cultured species (Wendy and Kevan, 1992).

Most of the shrimp production is depend on the genetics of wild populations. Culturing larvae in hatcheries where good rearing conditions and antibiotics prevent diseases but, at the same time, reduce the ability of the larvae to resist unfavorable environmental conditions. Thus an alternative method to antibiotic treatment is the application of probiotics.

1.2.2 Constraints in Black Tiger Shrimp Farming

To date, invertebrate immune systems have not been shown to produce antibodies. Thus people, researchers, consider viral diseases to be the main cause of the industry's collapse. However, there are many other factors which contributed to the shrimp culture recession in the past and constrain the sustainable development of the industry in the future (Rengpipat *et al.*, 1998a).

People were often being able to control these diseases relatively easily by applying antibiotics or taking technical measures. However, those measures were not able to control recent epizootics and diseases spread so quickly that, if a few shrimp were found dead or diseased, there is a high chance where whole pond would be dead or dying.

Due to disease infection that affected on black tiger shrimp, *P. monodon* worldwide in the past, many of the farmers turn to culture of another species of penaeids shrimp especially the white leg shrimp, *Litopenaeus vannamei*. They no longer want to invest their money on something that unsustainable and risky business.

Morever, many shrimp ponds were concentrated near estuaries where polluted water drains directly, and many shrimp farms suffer heavy losses from this pollution. Although the government has been strengthening the protection of the environment, the pollution from industry waste water and sewage is still a big problem for shrimp farming. Another cause of environmental problems is pollution from the shrimp farming industry itself by draining a large amount of shrimp feces and waste feed are drained into coastal waters with the effluent and this result in eutrophication.

1.3 Significance of study

During the past previous year, black tiger shrimp farming particularly affected by an epidemic of viruses like monodon baculovirus (MBV), white spot syndrome virus (WSSV), yellow head virus (YHV) and infectious hypodermal and hematopoietic

necrosis virus (IHHNV). Several disease outbreaks were also due to vibriosis, including *Vibrio alginolyticus*, *V. damsela*, *V. harveyi*, and *V. parahaemolyticus* (Song *et al.*, 1993; Lee *et al.*, 1996; Liu *et al.*, 1996; Sung *et al.*, 2001). These diseases have been reported to be associated with increases of *Vibrio* populations of cultured pond waters (Sung *et al.*, 2001).

The search for technological solutions to problems related to high density aquaculture practices has resulted in a proliferation of often unproven and potentially dangerous solution. Products and procedures such as chlorination, antibiotics, and even toxic insecticides are touted as cures for problems in hatcheries and farms. Although some of these products can improve the culture environment or exclude disease carries, misuse has shown to compounds existing problems (Balcazar *et al.*, 2006).

The solution lays in the field of microbial ecology, not in the field of pharmacology, i.e, in developing new antibiotics or vaccines (Moriarty, 1997). The use of effective microorganisms such as probiotics seems an alternative way to displace pathogenic bacteria by competitive process is a better remedy than administering antibiotics.

1.4 Objectives of study

In this study, an attempt was made to identify the affects of this probiotics on *P. monodon* by enhancing influence of *Bacillus* sp. into the rearing treatment tanks. Thus these are the objectives of this study:

1. To improve the survival and growth performance of post larvae with probiotics.
2. To determine the effects of probiotics on the post larvae after challenging with *Vibrio* sp.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction to Black Tiger Shrimp, *Penaeus monodon*

2.1.1 Classification of Black Tiger Shrimp

Kingdom : Animalia

Phylum : Arthropoda

Subphylum : Crustacea

Class : Malacostraca

Order : Decapoda

Suborder : Dendrobranchiata

Family : Penaeidae

Genus : Penaeus

Species : *Penaeus monodon*

FAO name : Black Tiger Shrimp

Local name : Udang harimau

2.1.1 Black Tiger Shrimp Life Cycle

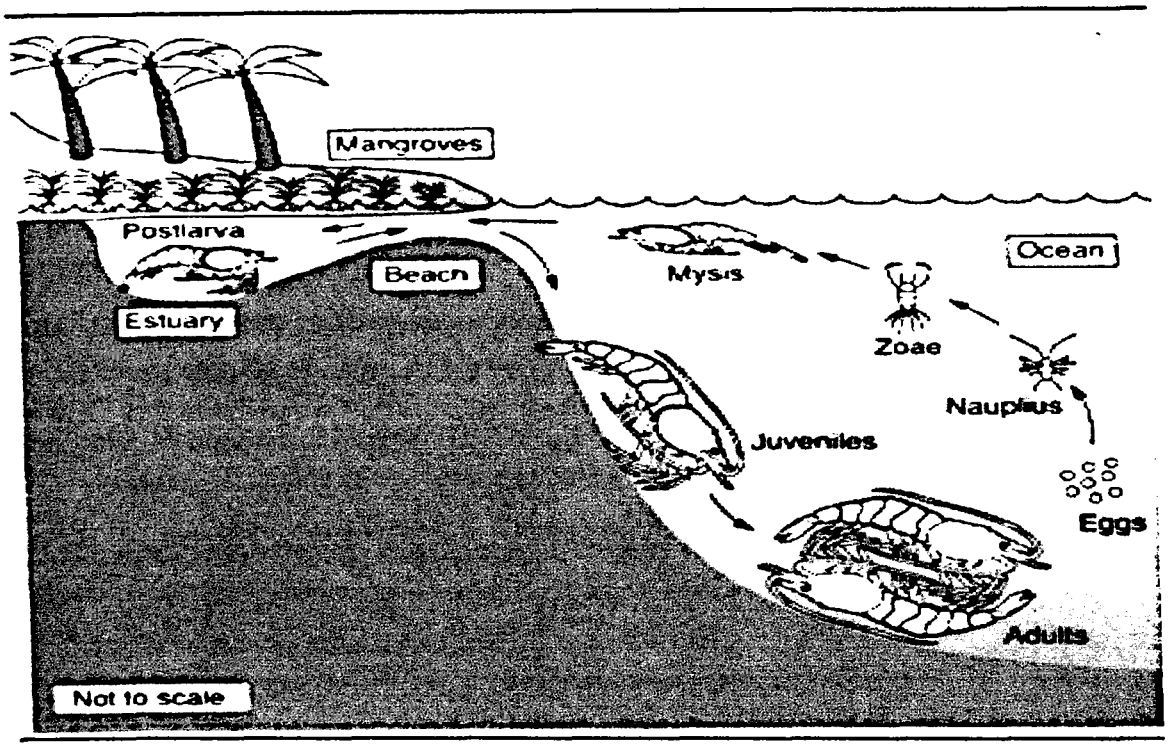

Figure 2.1 A typical life cycle of a marine shrimp (Rosenberry, 1999).

Start with some introduction of this subtopic. The shrimp will migrate up to several kilometers offshore to spawn. After mating took place, it will be produce a number of eggs and normally they will hatch between 15h to 20h. Most of the spawning area located in the favorable current for natural nursery stage. Young shrimp will spend several months in the estuary area before they migrate back to the ocean (Rosenberry, 1999).

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