

THE EFFECTS OF DIFFERENT SALINITIES
LEVEL ON HATCHING RATE OF TIGER
GROUPEP, *Epinephelus fuscoguttatus*
EGGS

CHENG ZI YANG



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**THE EFFECTS OF DIFFERENT SALINITIES ON HATCHING RATE OF
TIGER GROUPER (*Epinephelus fuscoguttatus*) EGGS**

CHENG ZI YANG



**THIS THESIS IS SUBMITTED AS A PARTIAL REQUIREMENT TO
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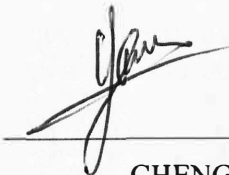
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DECLARATION

I declare that dissertation is the result of my own independent work and original writing, otherwise stated.

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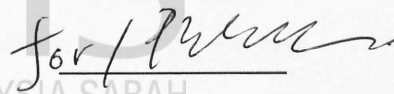
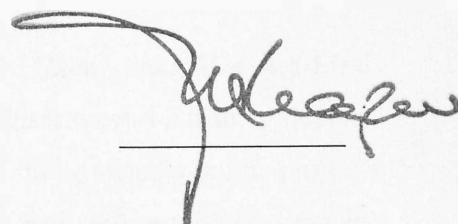
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AUTHENTICATION**AUTHENTICATED BY MEMBERS OF DISSERTATION COMMITTEE.**

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ABSTRACT

In order to solve the problem of low hatching rate of tiger grouper, *E. fuscoguttatus* in artificial seed production, an experiment based on eggs hatching rate in different salinity levels was done. The salinity levels used in the experiment were 0, 5, 10, 15, 20, 25, 30, and 35 ppt. The experiment was conducted in 500ml beaker. The eggs used in the experiment were collected through natural spawning of tiger grouper in the UMS hatchery. Collected eggs were transferred into beaker and acclimatisation process was done to prevent salinity shock to the eggs. At the water temperature of 28°C, tiger grouper eggs did not hatch at the salinity of 0 and 5 ppt. Under the same temperature, the hatching rate of tiger grouper eggs at the salinity of 10 ppt was 11.33% ± 12.56. Hatching rate of 21.0% ± 16.82 and 41% ± 16.82 was achieved at the salinity of 15 and 20 ppt respectively. For the salinity level at 25 ppt, the hatching rate was 48.0% ± 15.07. Hatching rate reached 93.33% ± 3.27 for the salinity level of 30 ppt. The hatching rate reached the highest at the salinity level of 35 ppt which was 98.0% ± 2.53. Better hatching rate can be achieved by culturing tiger grouper eggs in high salinity range from 30 – 35 ppt.

ABSTRAK

Bagi menyelesaikan masalah kadar penetasan telur kerapu harimau, *E. fuscoguttatus* yang rendah dalam pengeluaran benih, satu eksperimen berasaskan kadar penetasan dalam tahap kemasinan yang berlainan telah dijalankan. Tahap kemasinan yang telah digunakan adalah 0, 5, 10, 15, 20, 25, 30, dan 35 ppt. Eksperimen ini dijalankan dengan menggunakan bikar berisipadu 500 ml. Telur yang digunakan dalam eksperimen diperolehi dari hatcheri UMS melalui kaedah pengeluaran telur secara semula jadi oleh induk kerapu harimau. Telur kerapu harimau ini kemudiannya diletakkan dalam bikar dan proses penurunan kemasinan air secara perlahan-lahan dilakukan untuk mengurangkan kejutan kemasinan terhadap telur kerapu harimau. Pada suhu 28°C, tiada berlakunya penetasan telur pada tahap kemasinan 0 dan 5 ppt. Pada suhu yang sama, kadar penetasan telur kerapu harimau pada tahap kemasinan 10 ppt ialah 11.33% ± 12.56. Kadar penetasan mencatat 21.0% ± 16.82 pada tahap kemasinan 15 ppt. Kadar penetasan yang dikira mencapai 41.0% ± 16.82 pada kemasinan 20 ppt. Untuk tahap kemasinan 25 ppt, kadar penetasan ialah 48.0% ± 15.07. Kadar penetasan mencapai 93.33% ± 3.27 untuk kemasinan 30 ppt. Kadar penetasan mencapai tertinggi pada tahap kemasinan 35 ppt iaitu 98.0% ± 2.53. Untuk mencapai kadar penetasan yang bagus, telur kerapu harimau dinasihati agar menyimpan dalam air yang kemasinannya tinggi iaitu dari 30 hingga 35 ppt.

CONTENTS

	Page
DECLARATION	ii
AUTHENTICATION	iii
ACKKWOLEDGEMENTS	iv
ABSTRACT	v
ABSTRAK	vi
LIST OF CONTENTS	vii
LIST OF FIGURE	ix
LIST OF TABLE	x
LIST OF PHOTO	xi
LIST OF SYMBOL	xii
CHAPTER 1 INTRODUCTION	1
1.1 Vision of Aquaculture in Malaysia	1
1.2 Tiger Grouper (<i>Epinephelus fuscoguttatus</i>)	3
1.3 Constraints in <i>Epinephelus fuscoguttatus</i> Seed Production	5
1.4 Objectives of Study	6
CHAPTER 2 LITERATURE REVIEW	7
2.1 Taxonomy, Morphology, and Fish Culture of Tiger Grouper (<i>Epinephelus fuscoguttatus</i>)	7
2.2 Hormone-induced spawning	9
2.3 Eggs Development Stages	10
2.4 Effects of Salinity on Eggs	14
CHAPTER 3 MATERIALS AND METHODS	16
3.1 Brood Fish Management	16
3.2 Experiment Design and Set Up	17
3.2.1 Natural Spawning	21
3.2.2 Artificial Eggs Collection	21

CHAPTER 4 RESULTS	24
4.1 Hatching Rate	24
4.2 Egg Buoyancy	26
CHAPTER 5 DISCUSSIONS	27
5.1 Hatching Rate	27
5.2 Egg Buoyancy	30
CHAPTER 6 CONCLUSION	31
REFERENCES	33



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LIST OF TABLE

Table No.		Page
3.1	How formula is used to get the salinity levels that needed.	19



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LIST OF FIGURE

Figure No.	Page
2.1 6 stages of classification.	13
2.2 Egg development of tiger grouper.	14
3.1 Chemical compositions in sea salt	20
4.1 Average hatching rate of tiger grouper.	25
4.2 Division of beaker	27
4.3 Egg buoyancy of tiger grouper in different salinities	27



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LIST OF PHOTO

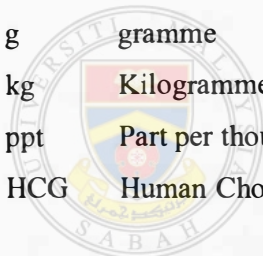
Photo No.	Page
1.1 Tiger grouper (<i>Epinephelus fuscoguttatus</i>)	4
3.1 Apparatus used for preparing water with salinity of 35 ppt	21
3.2 Hormone injection for female brood fish	23



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LIST OF SYMBOL

°C	Degree Celcius
%	Percent
DO	Dissolve Oxygen
L	Liter
SD	Standard deviation
IU	International Unite
mg	Miligramme
ml	Mililitre
g	gramme
kg	Kilogramme
ppt	Part per thousand
HCG	Human Chorionic Gonadotrophin hormone



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CHAPTER 1

INTRODUCTION

1.1 Vision of Aquaculture in Malaysia

Malaysia is located at the centre of Southeast Asia and covers an area of 127,000 sq.miles (330,200 km²). Malaysia is divided into two main regions: Peninsular Malaysia, which lies south of Thailand, and East Malaysia, which can be found south of Philippine on the island of Borneo. Hence about three quarter of its population of 23 million live in the Peninsular Malaysia. Being surrounded by sea, Malaysia has a coastline of 4800 km. Along this coastline the mangrove forest covers an area of about 641,000 ha with 57% of it located in Sabah, 26% in Sarawak and only 17% is in Peninsular Malaysia. (Mohd. Fariduddin, 2006)

Beyond the shore are over two hundred islands with warm clear waters and teeming marine life which have delighted underwater adventurers. On climate side, an average temperature range from 21⁰ C to 32⁰ C. Humidity in Malaysia is high. Rain tends to occur between November to February on the east coast of Peninsular Malaysia, on the western Sarawak, and north-eastern Sabah. On the west coast of Peninsular Malaysia the rainy

seasons is April to May and October to November. With its favorable climate supported by a vast resources Malaysia has a great potential for aquaculture development. (Mohd. Fariduddin, 2006)

Aspiring to become a develop country status by the year 2020, Malaysia recognises the significance of sustainable aquaculture as an integral part of efforts to develop its natural resources. On the way, Malaysia is putting up effort to increase its aquaculture production. An area that is given attention is shrimp and marine finfish aquaculture industry. Various institutions and government agencies had been given the task to commercialise this sector, get involve in research, training and development. On the other hand, mindful of the rising labor shortage in Malaysia, the government policy is to promote capital intensive large scale commercial shrimp aquaculture farms. Farms are encouraged to operate on an integrated and self-sustaining basis. Fry and feed production, processing and packaging, as well as marketing, are built into these vertically integrated systems. In achieving these Malaysia as well encouraged partnership. The government also interested in attracting foreign capital and appropriate know-how whenever is available to develop this sector through environment friendly technologies. (Mohd. Fariduddin, 2006)

1.2 Tiger Grouper

Based on the records in the World Fish Base (www.fishbase.org), tiger grouper, scientifically known as *Epinephelus fuscoguttatus*, is from the Family *Serranidae*, subfamily *Epinephelinae*, Order *Perciformes*, Class *Actinopterygii*, Genus *Epinephelus*, and species *fuscoguttatus*. This species are one of the larger species from the subfamily of *Epinephelinae*. This species has lots of name that varies from one country to another country. But this species is commonly known as tiger grouper or brown-marble grouper in Australia or kerapu harimau in Malaysia. This species is one of the favorite culture species in Malaysia.



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This species are reported can reached about 120 cm in total length and 11 kg in body weight. *Epinephelus fuscoguttatus* can found throughout the Indo-Pacific area, from the Red Sea and East Africa to Samoa and the Phoenix Islands, north up to Japan, south to Australia. It is a predatory fish that usually habit in the reef-associated marine area with the depth range around 1 m to 60 m (Stachels, 1996).

Mostly, *E. fuscoguttatus* occurs in lagoon pinnacles, channels, and outer reef slopes. It is also could be seen in coral-rich areas with clear waters. For the *E. fuscoguttatus* juveniles, it usually can be found among the sea grass beds. It feeds on

fishes, crabs, and cephalopods. Principally, *E. fuscoguttatus* start to be active at dusk (Tucker, 1999).

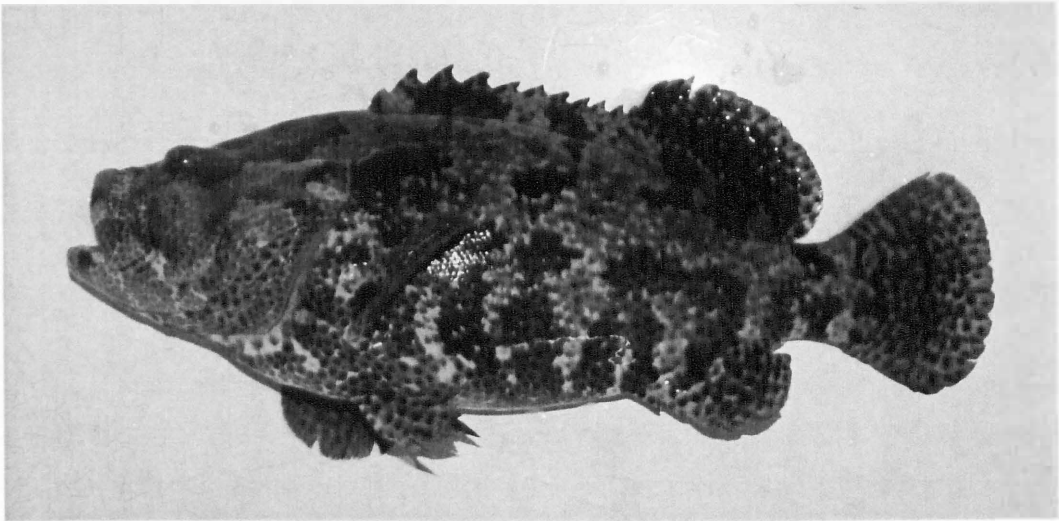


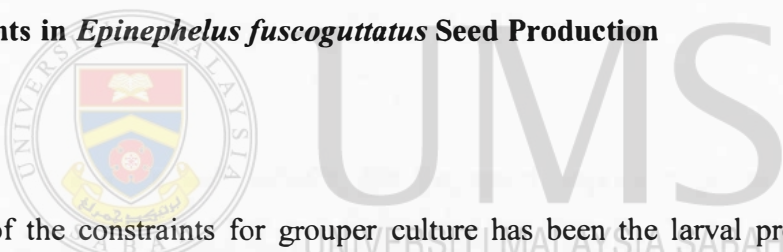
Photo 1.1: Tiger Grouper (*Epinephelus fuscoguttatus*)

This species is considered as a medium priced grouper but it has a very good price in Hong Kong especially live fish markets, which could reach HK\$237.50/kg to HK\$437.40/kg during year 1999 (Wilkinson and Cajilig, 2002). In Malaysia, this species also can sell from RM 80.00/kg to RM130.00/kg in seafood restaurant, but this price is very depending on season and occasion. The price will rise very high during the Chinese New Year.

This species is a very important aquaculture species due to its rapid growth, efficient feed conversion ratio, desirable taste and high market value. (Chen and Tsai, 1994; Millamena, 2002). The juvenile of this species have a high demand by the aquaculture farmers in Southeast Asia because of their good survival and rapid grow in

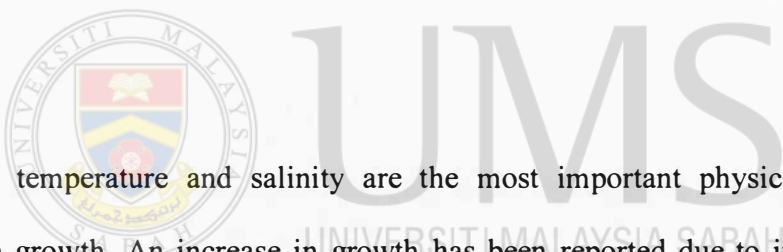
culture. Juveniles are sold in Hong Kong reef fish food market, the most common at 25–50cm length (Lee and Sadovy, 1998). The seeds production is easy under the artificial control. However the process to grow the seed is difficult. It is reported that they have had some problems with post-hatch larvae mortalities (Sadovy, 2000). Fry were grown from naturally spawned eggs (Nazar and Higuchi, 1980) but mortality is high. Besides seeds can be produce under artificial control but most of the Malaysian like to collect the seeds from the wild. Fry are collected mainly from the wild for cage culture in Peninsular Malaysia.

1.3 Constraints in *Epinephelus fuscoguttatus* Seed Production



One of the constraints for grouper culture has been the larval production (Lim, 1993). In larviculture of marine fishes, early mortality is one of the most critical matters. Early mortality of fish larvae is determined by both biotic and abiotic factors. Biotic factors such as predation, starvation, disease, and transportation to unprofitable physical conditions of water. For the abiotic factors such as DO, pH, temperature, and salinity (Houde, 1987). For this species, there are no data for the effects of temperature and salinity on hatch rate either on embryos or on larvae during yolk sac absorption up to first feeding (Gracia-Lo´pez *et al.*,2004).

Temperature and salinity influence marine fish eggs and larval physiology, having a direct effect on growth and survival (Holliday, 1969; Alderdice and Forrester, 1968). Water temperature influences hatching rates (Hart and Purser, 1995), larval sizes at hatch (Hansen and Falk-Petersen, 2001), time for yolk sac absorption (Pauly and Pullin, 1988), energy reserve take-up efficiency (Heming, 1982), and larval growth and survival (Akatsu *et al.*, 1983). Salinity affects hatching rate and egg diameter (Holliday, 1969), larval survival (Lee and Menu, 1981), yolk consumption efficiency (Swanson, 1996), and growth (Murashige *et al.*, 1991). Salinity also affects egg buoyancy, which has been studied by Craik and Harvey (1987) for a great number of species and has an important effect on egg survival in commercial hatcheries (Hart and Purser, 1995).



Water temperature and salinity are the most important physical factors that influence fish growth. An increase in growth has been reported due to increased water temperature for Nassau grouper (Ellis *et al.*, 1997), and several fish species. Lower salinities than seawater have also been found to affect fish, leading to increased growth (Chua & Teng, 1980; Alliot *et al.*, 1983)

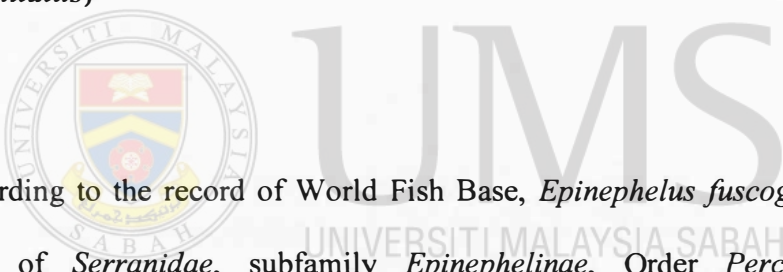
Since the salinities factors bring a great effect toward so many aspects in seed production which I intend to study the effect of different salinities level on the egg hatching rate of *Epinephelus fuscoguttatus*. The objectives of this study are as following:

1. To determine the best salinity for the optimum hatching rate
2. To determine the effects of different salinity levels on eggs hatching rate

CHAPTER 2

LITERATURE REVIEW

2.1 Taxonomy, Morphology, and Culture of Tiger Grouper (*Epinephelus fuscoguttatus*)



According to the record of World Fish Base, *Epinephelus fuscoguttatus* is from the Family of *Serranidae*, subfamily *Epinephelinae*, Order *Perciformes*, Class *Actinopterygii*, and Genus *Epinephelus*. From the record, this species has been reported can be reached until 120 cm in total length and 11 kg in body weight. In the record, it also stated that this species could be found in the reef-associated marine area, lagoon pinnacles, channels, and outer reef slopes with the dept range around 1 m to 60m.

In the record from World Fish Base, this species, *Epinephelus fuscoguttatus*, morphologically has totally of 11 dorsal spines, 14 to 15 dorsal soft rays, 3 anal spines, and 8 anal soft rays. For juveniles that less than 8 cm, it usually have small hexagonal spots on head and body. The spots will become larger posterior and also can be found on

vertical fins. Most tiger grouper mature within 2 to 6 years. It can be reared in cages, ponds, and tanks, with the water temperature around 28°C to 32°C. Usually it fed on trash fish started from the juvenile stage (Ch'ng, 2007).

From the records of World Fish base, *E. fuscoguttatus* has the ability to doubling the population in 1.4 to 4.4 years. *Epinephelus fuscoguttatus* has totally 11 dorsal spines, 14 to 15 dorsal soft rays, 3 anal spines, and 8 anal soft rays. For juveniles that less than 8 cm, it usually have small hexagonal spots on head and body. The spots will become larger posteriorly and also can be found on vertical fins (World Fish Base, <http://www.fishbase.com>).



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In University Malaysia Sabah hatchery, the larvae of Tiger grouper will be kept in green water (*Nannochloropsis*, *Tetraselmis*, *Chorella* spp.) for duration of 35 to 40 days. This is because during this stage, the larvae are very sensitive to sunlight. Besides as the protection for the larvae, the phytoplankton within the green water as can acts as foods for the rotifers when the larvae are fed with rotifer. Survival rate and growth rate tends to be increase if copepod or mixed zooplanktons are included in the diet. Enriched *Artemia* can be a staple food beginning at 10 to 30 days, but their density should be controlled to minimise gorging (Ch'ng, 2007).

2.2 Hormone-induced Spawning

Hormone induced spawning is a very important technique that wisely use in the aquaculture sector. This technique has been used for more than 50 years to make the brood stocks that do not perform spawning in captivity. Besides that, this technique can also use to gain control of fry production.

Preferably broodstock maintained under optimum conditions, such as environmental stimuli, water quality, nutrition, pH and water temperature should spawn in captivity; however, brood stock capture from wild often fail to progress through oocyte maturation and ovulation due to differences between natural environment conditions and the hatchery (Zohar, 1988).

Broodstocks that already treated with hormone can sub divided into two groups; the first groups where the brood stocks can spawn naturally in the captivity after the hormone treatment while the other groups is the brood stocks have to undergo the process of artificial stripping. In this process of artificial stripping, eggs can be stripped through final maturation in which germinal vesicle degenerates, the first meiotic division is complete, the second meiotic division begins and is arrested, and the hydrated oocytes are released into the ovarian lumen (Wallance *et al.* 1987).

However, there are some considerations when manually stripping a fish to obtain the eggs. One needs to know when ovulation occurred and at what point post-ovulation can the highest quality eggs be collected. Besides knowing when stripping can be done, the water temperature also is a very important factor in artificial seed production. This is due to each species of fish has an appropriate temperature range for spawning. When an animal is induced to spawn within that temperature range, small differences in temperature may delay or accelerate ovulation. Because most fish are poikilotherms, the rate of a physiological reaction, including the response to an exogenous ovulation-stimulating hormone is controlled by temperature. (Phelps *et al.*, 2007).

Timing of ovulation is an essential component in stripping fertile eggs from brood stock. Eggs stripped too early are immature, infertile oocytes, while eggs retained in a female after ovulation began a process of over ripening and degradation (Bromage *et al.*, 1994). Over ripening continues from the time eggs are stripped from female to the time sperm is added for fertilization. Immature and over ripe eggs stripped from brood stock can cause significantly lower fertilization rates and lower larval survival rates (Kjorsvick *et al.*, 1990).

2.3 Eggs Development Stages

When the milt from male and eggs from female are mixed, fertilisation will occur. Once the fertilization is complete, rapid cell division will occur. During this period, cleavage,