

THE EFFECT OF EXOGENOUS THROXINE ON THE
MORPHOLOGY AND DIGESTIVE SYSTEM DEVELOPMENT
DURING THE EARLY LIFE STAGES OF TIGER
GROUPEP (*Epinephelus fuscoguttatus*)

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MASTER OF SCIENCE
UNIVERSITI MALAYSIA TERENGGANU
MALAYSIA

2011

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**Thesis Submitted in Fulfillment of the
Requirement for the Degree of Master of
Science in the Institute of Tropical Aquaculture
Universiti Malaysia Terengganu**

JUNE 2011

DEDICATION

This thesis is dedicated to:

- 1. My lovely wife Ernayanti Utami and my son Azril Ahmad Alfarizzi, my Parents Umedi and Siti juhriyah; Mukhsin Syamsudin and Suryati, my brothers Akbar Zainuddin and family; Kholid Rizaluddin and Family; Hendra Wahyu Saputra and Mohammad Daffa Faturrahman. For their support and time they gave to me in entire my life.*
- 2. My Supervisor Assoc. Prof. Dr. Abol Munafi Ambok Bolong and Prof. Dr. Mohd. Effendy Abd. Wahid for his support during my study in Malaysia.*

Abstract of thesis presented to the Senate of Universiti Malaysia Terengganu in fulfillment of the requirement for the degree of Master of Science.

EFFECT OF EXOGENOUS THYROXINE ON MORPHOLOGY DEVELOPMENT AND DIGESTIVE SYSTEM DURING EARLY LIFE STAGE OF TIGER GROUPER (*Epinephelus fuscoguttatus*)

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June, 2011

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Tiger grouper (*Epinephelus fuscoguttatus*) is one of the most commercially important grouper species and commanding high prices. *Epinephelus fuscoguttatus* aquaculture has been established since the past 10 years but its seeds production is problematic and unstable. The average survival rate is low and early mortality often occurs especially between four and nine days after hatching. A study on the effects of exogenous thyroxine (T_4) on the morphological development and digestive system during early life stage of *E. fuscoguttatus* was carried out to establish the knowledge on the role of thyroid hormone in improving the growth, survival and the digestive tract developmental changes associated with food assimilation processes. The objectives of this study are; (i) To observe the effect of exogenous thyroxine on growth and survival rate of *E. fuscoguttatus* larvae. (ii) To study the effect of exogenous thyroxine on the morphological development and ontogeny of

digestive tract in early life stage of *E. fuscoguttatus*. This study was conducted at the Research Institute for Mariculture Gondol-Bali Indonesia and Institute of Tropical Aquaculture of Universiti Malaysia Terengganu.

Newly hatched larvae of *E. fuscoguttatus* were immersed with 0.2 ppm and 0.1 ppm of T₄ for 2 hours and reared until 40 DAH. One group was reared without T₄ treatment as a control. In the first experiment, administrations of T₄ have significantly increased the total length and growth rate of larvae. The highest total length and growth rate were in the larvae treated with 0.2 ppm of T₄ (TL: 32.96 ± 2.28 mm and GR: 0.78 ± 0.057 mm day⁻¹, respectively), followed by larvae treated with 0.1 ppm of T₄ (TL: 30.55 ± 2.04 mm and GR: 0.72 ± 0.052 mm day⁻¹) and the control group (TL: 28.15 ± 2.05 mm and GR: 0.66 ± 0.051 mm day⁻¹). There was no significant difference in the survival rate of treated and untreated *E. fuscoguttatus* larvae. Thyroxine treatment also has significantly increased the concentration of total T₄ of the treated larvae compared to the control group. Total T₄ concentration at the end of experiment of larvae treated with 0.1 and 0.2 ppm of T₄ were 1.54 ± 0.06 µg dL⁻¹ and 1.53 ± 0.01 µg dL⁻¹ respectively, while the total T₄ concentration of the control group was 1.50 ± 0.01 µg.dL⁻¹.

In the second experiment, morphological observation and standard histological protocols were conducted to observe the digestive

system of *E. fuscoguttatus* larvae. The exogenous T₄ treatment stimulated notochord flexion, development of fin rays, appearance of melanophores in dorsal part of larvae, reduction of second dorsal fin spine and pelvic fin spine and accelerated the metamorphosis of *E. fuscoguttatus* larvae compared to the control group larvae. Juvenile and adult stage of the treated larvae were detected earlier at 19 – 40 DAH and 34 – 40 DAH respectively in treated larvae compared to control larvae at 22 – 40 DAH and 37 – 40 DAH respectively. Histological observation showed that several digestive tracts organs of treated larvae were observed earlier compared to control groups, such as goblet cells, tongue, gill raker, fundic and cardiac stomach, pharyngeal tooth and gastric gland. Acidophilic supranuclear and lipid vacuoles were observed more in the rectum of larvae treated with thyroxine compared to the control groups.

In conclusion, exogenous T₄ treatment increased the total length, growth rate and concentration of total T₄; accelerated the metamorphosis; stimulated the digestive tracts development and increased digestive activity of *E. fuscoguttatus* larvae.

Abstrak tesis yang dikemukakan kepada Senat Universiti Malaysia Terengganu sebagai memenuhi keperluan untuk ijazah Sarjana Sains

KESAN TIROKSINA EKSOGENUS KE ATAS PERKEMBANGAN MORFOLOGI DAN SISTEM PENCERNAAN PADA PERINGKAT AWAL HIDUP KERAPU HARIMAU (*Epinephelus fuscoguttatus*)

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Kerapu Harimau (*Epinephelus fuscoguttatus*) merupakan salah satu spesies kerapu yang penting secara komersial dan mempunyai harga yang tinggi. *Epinephelus fuscoguttatus* telah berjaya ditenak sejak 10 tahun yang lalu namun pengeluaran benihnya masih lagi bermasalah dan tidak stabil. Purata kadar kemandirian benihnya adalah rendah dan kematian awal sering berlaku terutamanya antara hari keempat dan kesembilan selepas menetas. Satu kajian mengenai kesan tiroksina eksogenus (T_4) ke atas perkembangan morfologi dan sistem pencernaan pada peringkat awal hidup *E. fuscoguttatus* telah dijalankan untuk mendapatkan maklumat mengenai peranan hormon tiroid dalam meningkatkan pertumbuhan, kemandirian dan perkembangan saluran penghadaman yang berkait dengan proses asimilasi makanan. Objektif kajian ini adalah; (i) Untuk melihat kesan tiroksina eksogenus kepada pertumbuhan dan kadar kemandirian larva *E. fuscoguttatus*. (ii) Untuk mengkaji kesan

tiroksina eksogenus kepada perkembangan morfologi dan ontogeni saluran penghadaman pada peringkat awal kehidupan *E. fuscoguttatus*. Kajian ini telah dijalankan di Institut Penyelidikan Marikultur Gondol-Bali, Indonesia dan Institut Akuakultur Tropika, Universiti Malaysia Terengganu.

Larva *E. fuscoguttatus* yang baru menetas telah direndam didalam 0.2 ppm dan 0.1 ppm T_4 selama 2 jam dan dipelihara sehingga hari ke 40 selepas menetas. Satu kumpulan larva telah dipelihara tanpa dirawat dengan T_4 sebagai kawalan. Di dalam eksperimen yang pertama, rawatan T_4 telah meningkatkan dengan ketara panjang keseluruhan dan kadar kemandirian larva. Panjang keseluruhan dan kadar kemandirian tertinggi adalah pada larva yang telah dirawat dengan 0.2 ppm T_4 (TL: 32.96 ± 2.28 mm dan GR: 0.78 ± 0.057 mm hari⁻¹, masing-masing), diikuti dengan larva yang dirawat dengan 0.1 ppm T_4 (TL: 30.55 ± 2.04 mm dan GR: 0.72 ± 0.052 mm hari⁻¹) dan kumpulan kawalan (TL: 28.15 ± 2.05 mm dan GR: 0.66 ± 0.051 mm hari⁻¹). Tiada perbezaan yang ketara pada kadar kemandirian larva *E. fuscoguttatus* yang dirawat dan tidak dirawat. Rawatan tiroksina juga telah meningkatkan dengan ketara kadar jumlah kepekatan T_4 pada larva yang dirawat berbanding kumpulan kawalan. Jumlah kepekatan T_4 pada akhir eksperimen untuk larva yang dirawat dengan 0.1 ppm dan 0.2 ppm T_4 adalah masing-masing 1.54 ± 0.06 $\mu\text{g dL}^{-1}$ dan 1.53 ± 0.01 $\mu\text{g dL}^{-1}$, manakala jumlah kepekatan T_4 untuk kumpulan kawalan adalah 1.50 ± 0.01 $\mu\text{g.dL}^{-1}$.

Didalam eksperimen kedua, pemerhatian morfologi dan protokol histologi standard telah dijalankan untuk memerhatikan sistem pencernaan larva *E. fuscoguttatus*. Rawatan T₄ eksogenus telah merangsang lengkungan notokod, perkembangan sirip, kemunculan melanofor pada bahagian dorsal larva, pengurangan sirip dorsal kedua dan duri sirip pelvik serta mempercepatkan metamorfosis larva *E. fuscoguttatus* berbanding larva kumpulan kawalan. Peringkat juvenil dan dewasa dikesan seawal hari ke 19 hingga 40 DAH dan hari ke 34 hingga 40 DAH pada larva yang dirawat dengan tiroksina eksogenus berbanding larva kawalan pada hari ke 22 hingga 40 DAH dan hari ke 37 hingga 40 DAH masing-masing. Pemerhatian histologi menunjukkan bahawa beberapa organ pada saluran pencernaan larva yang dirawat dapat dilihat lebih awal berbanding kumpulan kawalan seperti sel goblet, lidah, insang, perut fundik dan kardiak, gigi faringeal dan pundi gastrik. Supranuklear asidofilik dan vakul lipid dapat dikesan lebih banyak pada rektum larva yang dirawat dengan tiroksina berbanding kumpulan kawalan.

Sebagai kesimpulan, rawatan T₄ eksogenus meningkatkan panjang keseluruhan; kadar kemandirian dan jumlah kepekatan T₄; mempercepatkan metamorfosis; merangsang perkembangan salur pencernaan dan meningkatkan aktiviti pencernaan pada larva *E. fuscoguttatus*.