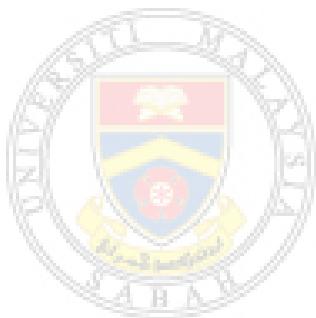


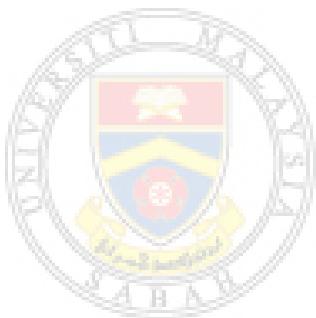
**EFFECT OF DIETARY DHA AND CAROTENOID
ON REPRODUCTIVE PERFORMANCE OF
PURPLE MANGROVE CRAB (*Scylla*
trannebarica) BROODSTOCK**



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THIEN FUI YIN
UNIVERSITI MALAYSIA SABAH

**BORNEO MARINE RESEARCH INSTITUTE
UNIVERSITI MALAYSIA SABAH
2019**

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PURPLE MANGROVE CRAB (*Scylla*
trannequebarica) BROODSTOCK**



THIEN FUI YIN
UMS
UNIVERSITI MALAYSIA SABAH

**THESIS SUBMITTED IN FULFILLMENT FOR
THE DEGREE OF MASTER OF SCIENCE**

**BORNEO MARINE RESEARCH INSTITUTE
UNIVERSITI MALAYSIA SABAH**

2019

UNIVERSITI MALAYSIA SABAH

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DECLARATION

I hereby declare that the material in this thesis is my own except for quotations, excerpts, equations, summaries and references, which have been duly acknowledged.

06 AUGUST 2018

THIEN FUI YIN

MY1421040T



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CERTIFICATION

NAME : THIEN FUI YIN

MATRIC NO. : MY1421040T

TITLE : EFFECT OF DIETARY DHA AND CAROTENOID ON
REPRODUCTIVE PERFORMANCE OF PURPLE
MANGROVE CRAB (*Scylla tranquebarica*)
BROODSTOCK

DEGREE : MASTER OF SCIENCE (AQUACULTURE)

VIVA DATE : 24 JANUARY 2019



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CERTIFIED BY
UNIVERSITI MALAYSIA SABAH

Signature

1. Main Supervisor

Assoc. Prof. Dr. Annita Yong Seok Kian

2. Co-Supervisor

Professor Dr. Saleem Mustafa

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ABSTRACT

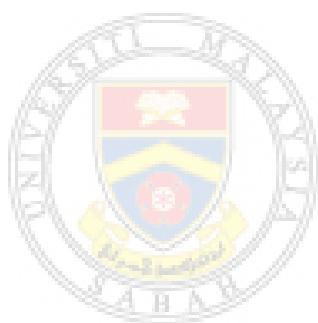
In mangrove crab seed production, it is important to provide the broodstock with maturation diet that can improve the reproductive performance. For the purple mangrove crab (*Scylla tranquebarica*), a dominant mangrove crab in Sabah, there are limited study on its maturation diet. Therefore, the present study was conducted to investigate the suitable maturation diet for the purple mangrove crab. Formulated diets (46% crude protein, 12% crude lipid) were prepared by adding astaxanthin (500 mg kg⁻¹) (FA), DHA (10 g kg⁻¹) (FD), both astaxanthin and DHA (FA+D) and without astaxanthin and DHA (Fcon). Natural food (NF) was comprised of trash fish (*Valamugil* sp.), shrimp (*Penaeid* sp.) and mangrove clam (*Polymesoda* sp.). Immatured female broodstock of initial body weight 129.07 ± 17.03 g and male 125.30 ± 14.12 g were cultured under recirculating water system for 50 weeks with the ratio of 1:5 (male:female). Reproductive performance of the broodstock was routinely monitored. Matured females (n=3) were identified and sacrificed for gonadosomatic index (GSI) estimation and biochemical analysis while others were allowed to spawn. The male broodstock (n=3) also sacrificed to determine the sperm viability. Newly hatched larvae (from one hatching) were measured (n=30), subjected to resistance tests (ammonia, dissolve oxygen, salinity and starvation) and reared until zoea II to determine the survival rate. The results revealed that the broodstock fed FA+D showed significantly higher maturation and spawning rate than broodstock fed Fcon, however, it showed no significant different with broodstock fed NF. Broodstock fed FA+D also showed significant better results than Fcon and NF in term of GSI, ovary colour intensity, oocyte diameter, molting rate, molt death syndrome (MDS) and male sperm viability. Broodstock given FD and FA diets resulted in significantly better results than NF group in term of GSI, oocytes diameter and sperm viability ($P<0.05$). However, the broodstock fed FD, FA and NF diets were not significant different to each other in term of maturation, spawning, molting, MDS and mortality ($P>0.05$). On the other hand, broodstock fed FA and FA+D diets showed significantly greater total carotenoid accumulation in the ovary than other treatments corresponding to the ovary colour intensity ($P<0.05$). However, total carotenoid accumulated in the female hepatopancreas was not significant different among the treatments ($P>0.05$). Broodstock fed FD and FA+D diets also showed significantly higher DHA accumulation in the ovary and hepatopancreas than those fed Fcon diet ($P<0.05$) but not significantly different with FA and NF groups ($P>0.05$). Larval from broodstock fed FA+D diet showed higher hatching rate, total number of larvae and significantly higher zoea II survival than Fcon group ($P<0.05$). Likewise, larval resistance tests showed that the larvae produced by broodstock fed FA+D diet showed significantly higher survival rate than Fcon and NF groups in all of the tests. In conclusion, formulated feed that supplemented with DHA and astaxanthin is a better maturation diet than the natural food for purple mangrove crab. The maturation diet provided also affected the early stage larval quality with best quality observed in larvae produced by the broodstock fed DHA and astaxanthin.

ABSTRAK

KESAN PEMAKANAN DHA DAN KAROTENOID TERHADAP PRESTASI PEMBIAKAN INDUK KETAM BAKAU UNGU (*Scylla tranquebarica*)

Dalam penghasilan benih ketam bakau, adalah penting untuk menyediakan induk dengan makanan kematangan yang dapat mempertingkatkan prestasi pembiakan dalam penghasilan benih. Untuk ketam bakau ungu (*Scylla tranquebarica*), ketam bakau dominan di Sabah, kurang penyelidikan dijalankan untuk mengkaji makanan kematangannya. Oleh itu, kajian ini telah dijalankan untuk mengkaji makanan kematangan yang sesuai untuk ketam bakau ungu. Rumusan makanan (46% protein mentah, 12% lipid mentah) telah disediakan dengan penambahan astazantin (500 mg kg⁻¹) (FA), DHA (10 g kg⁻¹) (FD), kedua-dua astazantin and DHA (FA+D) dan tanpa astazantin dan DHA (Fcon). Makanan semula jadi (NF) terdiri daripada ikan (*Valamugil* sp.), udang (*Penaeid* sp.) and lokan (*Polymesoda* sp.). Induk betina yang belum matang dengan berat badan awal 129.07 ± 17.03 g dan jantan 125.30 ± 14.12 g telah dikultur dalam sistem air kitaran selama 50 minggu dengan nisbah 1:5 (jantan:betina). Prestasi pembiakan induk dipantau secara berkala. Betina matang (n=3) telah dikenalpasti dan dikorbankan untuk menganggar index gonad (GSI) dan analisis biokimia sementara yang lain dibiarkan bertelur. Induk jantan (n=3) juga dikorbankan untuk menentukan peratus sperma hidup. Larva yang baru menetas (dari satu kali penetasan) diukur (n=30), diuji dalam ujian ketahanan (ammonia, oksigen terlarut, saliniti dan kelaparan) serta dijaga sehingga zoea II untuk menentukan kadar kemandiriannya. Keputusan memaparkan bahawa induk yang diberi makanan FA+D menunjukkan perbezaan bererti dalam peratus kematangan and bertelur berbanding dengan induk yang diberi makanan Fcon, walau bagaimanapun, tidak ada perbezaan bererti dengan induk yang diberi NF. Induk yang diberi makanan FA+D juga menunjukkan keputusan yang lebih baik dengan bererti berbanding kumpulan Fcon dan NF dalam GSI, warna ovarii, kadar salin kulit, sindrom kematian salin kulit (MDS) dan peratus sperma hidup. Induk yang diberi makanan FD dan FA menunjukkan keputusan yang lebih baik dengan bererti dalam GSI, diameter telur dan peratus sperma hidup berbanding kumpulan NF ($P<0.05$). Walau bagaimanapun, induk yang diberi makanan FD, FA and NF tidak menunjukkan perbezaan bererti dalam peratus kematangan, bertelur, salin kulit, MDS dan kadar kematian ($P>0.05$). Sementara itu, induk yang diberi makanan FA and FA+D menunjukkan perbezaan bererti untuk jumlah kandungan karotenoid terkumpul dalam ovarii berbanding dengan kumpulan lain selaras dengan warna ovarinya ($P<0.05$). Walau bagaimanapun, jumlah kandungan karotenoid terkumpul dalam hepatopancreas tidak menunjukkan perbezaan bererti antara semua kumpulan ($P>0.05$). Induk yang diberi makanan FD and FA+D pula menunjukkan perbezaan bererti dalam kandungan DHA terkumpul di dalam ovarii dan hepatopancreas berbanding dengan induk yang diberi makanan Fcon ($P<0.05$) tetapi tiada perbezaan bererti dengan kumpulan FA and NF ($P>0.05$). Larva berhasil daripada induk yang diberi makanan FA+D menunjukkan kadar penetasan dan jumlah larva yang lebih tinggi serta kadar kemandirian bagi zoea II yang bererti berbanding kumpulan Fcon ($P<0.05$). Begitu juga, keputusan dalam semua ujian ketahanan menunjukkan bahawa larva yang dihasilkan oleh induk FA+D menunjukkan kadar kemandirian yang bererti berbanding dengan kumpulan NF and Fcon. Kesimpulannya, makanan rumusan yang ditambah dengan DHA dan astazantin adalah lebih baik daripada makanan semula jadi bagi ketam bakau ungu. Makanan kematangan yang diberikan juga mempengaruhi kualiti peringkat awal larva dan kualiti larva terbaik telah

diperhatikan terhasil daripada kumpulan induk yang diberikan makanan DHA dan astazantin.



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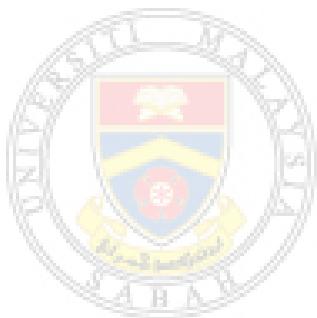
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LIST OF SYMBOLS

$^{\circ}\text{C}$	-	degree Celsius
$\%$	-	percentage
\pm	-	plus and minus
$\times 10^6$	-	times 1 million
$100x$	-	one hundred times
$400x$	-	four hundred times
I	-	one
II	-	two
III	-	three
IV	-	four
V	-	five



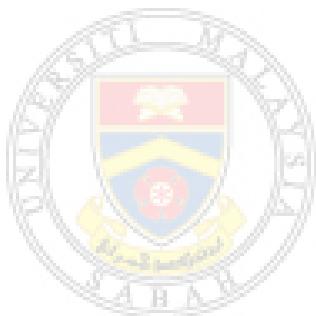
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AC	-	ash content
ANOVA	-	one way analysis of variance
ASTA	-	American Spice Trade Association
C14:0	-	myristic acid
C15:0	-	pentadecanoic acid
C16:0	-	palmitic acid
C16:1	-	palmitoleic acid
C17:0	-	heptadecanoic acid
C18:0	-	stearic acid
C18:1N9C	-	oleic acid
C18:1N9T	-	elaidic acid
C18:2N6C	-	linoleic acid
C18:3N3	-	α-linolenic acid
C20:4N6	-	arachidonic acid
C20:5N3	-	eicosapentaenoic acid
C22:1N9	-	erucic acid
C22:6N3	-	docosahexaenoic acid
CLC	-	crude lipid content
cm	-	centimetre
CMC	-	carboxymethylcellulose
DF	-	dilution factor
DHA	-	docosahexaenoic acid
DO	-	dissolved oxygen
E	-	east
EPA	-	eicosapentaenoic acid
<i>et al.</i>	-	and others
FAME	-	Fatty acid methyl ester
FC	-	fiber content
FID	-	flame ionization detector
g	-	gram
g kg ⁻¹	-	gram per kilogram
g/100g	-	gram per 100 gram

GC	-	gas chromatography
GSI	-	gonadosomatic index
H ₂	-	hydrogen gas
HR	-	hatching rate
HUFA	-	highly unsaturated fatty acids
ind L ⁻¹	-	individual per litre
ind ml ⁻¹	-	individual per millilitre
kg	-	kilogram
L	-	litre
L min ⁻¹	-	litre per minute
M	-	mole
m ²	-	metre square
MC	-	moisture content
MDS	-	molt death syndrome
mg kg ⁻¹	-	milligram per kilogram
mg L ⁻¹	-	milligram per litre
mL min ⁻¹	-	millilitre per minute
mm	-	millimetre
MR	-	molting rate
MR	-	mortality rate
MUFA	-	monounsaturated fatty acid
n	-	number of sample
N	-	north
N	-	nitrogen
N ₂	-	nitrogen gas
N	-	normality
n-3	-	omega 3 fatty acid
n-3/n-6	-	ratio of omega 3 fatty acid over omega 6 fatty acid
n-6	-	omega 6 fatty acid
ND	-	not detected
NFE	-	nitrogen free extract
nm	-	nanometre
pc	-	piece
ppm	-	parts per million

ppt	- parts per thousand
PTFE	- polytetrafluoroethylene
PUFA	- polyunsaturated fatty acid
RAS	- recirculating aquaculture system
RM	- Ringgit Malaysia
SD	- standard deviation
SFA	- saturated fatty acid
SPSS	- Statistical Package for the Social Sciences
SV	- sperm viability
TCC	- total carotenoid content
UV	- ultraviolet
ZSR	- zoea II survival rate
µm	- micrometer
µL	- microlitre



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