UNIVERSITI PUTRA MALAYSIA

STUDY ON THE DEVELOPMENT OF EMBRYO AND LARVA OF THE HUMPBACK GROUPER (CROMILEPTES ALTIVELIS VALENCEE) AND ITS FEEDING BEHAVIOUR

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By

USMAN BULANIN

Thesis Submitted in Fulfilment of the Requirement for the Degree of Doctor of Philosophy in the Faculty of Agriculture Universiti Putra Malaysia

February 2002

DEDICATION

To my parents Bulanin (alm.) and Hj. Mariawan,
my wife Misuharti and my child An-nisa Usman who gave me
supports and understandings during my study in Malaysia

Abstract of thesis submitted to the Senate of
Universiti Putra Malaysia in fulfilment of the requirement
for the degree of Doctor of Philosophy

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Chairman : Che Roos Saad, Ph.D
Faculty : Faculty of Agriculture

A series of experiments were conducted to determine 1) the embryonic and larval development, 2) feeding performance and 3) weaning of humpback grouper, *Cromileptes altivelis*, larva.

The above study was carried out from spawning of broodstocks to obtain the fertilized eggs till they hatched. The results showed that the mean diameter of humpback grouper egg was 828.69 ± 45.91 µm. The fertilized eggs hatched in 20 hour 10 minutes at the temperature of 27 to 28 °C. The hatched larvae had a mean length of 1.86 mm, and a mean height of 0.460 mm. The yolk sacs and oil droplets were totally utilized after 63 and 65 hour after hatching (HAH), respectively. Within one day, the larvae had an average length and height of 2.253 mm and 0.615 mm, respectively. At 49 DAH, the mean length and height increased to 27.153 mm and 9.190 mm respectively.

The spines on the larvae began to appear at day 7 to day 10. The dorsal spines reached their maximum length at day 28 to day 30, and the spines had a maximum mean length of 5668.30 μm, while the ventral spines had a maximum mean length of 4415.44 μm at day 25 – 28. Then the spines began to reduce and change to hard rays as the larvae grew.

The eyes pigments started to be visible at day 3 to day 4 after hatching (DAH). The larvae's mouths were opened for the first time at day 2 or 45 HAH. The relationship of mouth gape and eyes diameter was quadratic with an equation of MG = -02E-05Ed^2 + 0.3992Ed + 215.05 and R^2 = 0.96. Appearance of black spots at the caudal peduncle, dorsal fin and anal fin occurred between day 25 to day 30. These spots would increase in numbers and would spread towards the head and became perfected at day 40 to day 45, and they would look similar to the juvenile fish.

The teeth of humpback grouper fish larvae began to appear at day 15, but those teeth were still few in numbers and were short. The height and width of larvae teeth increased as the larvae grew. At day 30, the larvae teeth got closer with a mean height and width of 110.18 μm and 40.13 μm respectively.

Histologically, no fold occurred in the larvae digestive tract yet before they reach 5 days old. The incipient stomach began to be visible at day 30. The larvae's stomachs were perfected at 45 DAH. The liver began to be visible at day 5, while the pancreas was visible at day 10. The protease enzyme activity increased as the larvae grew.

Food was first found in the larvae's digestive tracts at day 3 or 66 HAH. The average number of total food found in the larvae's digestive tracts was 2 ind. larvae^{-1}. Rotifers with a mean size of 55.44 – 96 μm were first found in the larvae's digestive tracts. The larvae first consumed copepods with a mean size of 104.16 – 116.28 μm were by at day 6, while *Artemia* nauplii at day 15.

A 1-day old larva had a weight of 0.067 mg and its weight increased to141.114 mg at the end of the experimental with a growth rate of 2.88 mg day^{-1}. The percentage survival of larvae till 49 days was 3.03%.

The larvae that were fed with *Artemia* had a growth rate of 1.91 mg day^{-1}, and were significantly higher ($p < 0.01$) than larvae that were fed with artificial feed starting from day 35, 40 and 45. The growth rates of the larvae that were fed the artificial diet on day 35, 40 and 45 were 0.09 mg day^{-1}, 0.022 mg day^{-1} and – 0.02 mg day^{-1} respectively. The larvae that were given artificial feed at an early stage grew better than larvae fed on artificial feed at a later stage. Larvae fed on artificial feed on day 35 had a higher significant growth rate than the larvae fed with artificial feed on day 40. The mean survival of the larvae that were given *Artemia* was 21.34%, while larvae that were given the artificial feed at day 35, 40 and 45 were 8, 18 and 19.33%, respectively.

Abstrak tesis dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi syarat untuk mendapatkan ijazah Doktor Falsafah

KAJIAN PERKEMBANGAN EMBRIO DAN LARVA TERHADAP IKAN KERAPU TIKUS (*CROMILEPTES ALTIVELIS* VALENCEE) DAN CORAK MAKANNYA

By

USMAN BULANIN

Februari 2002

Pengerusi : Che Roos Saad, Ph.D
Fakulti : Fakulti Pertanian

Satu siri penyelidikan telah dijalankan untuk mengkaji 1) Perkembangan embrio dan larva, 2) Corak pemakanan 3) Permulaan masa untuk makanan buatan.

Kesemua kajian telah dijalankan mulai dari peneluran oleh induk sehingga penetasan. Keputusan menunjukkan purata diameter telur ikan kerapu tikus ialah 828.69 ± 45.91 μm. Penetasan telur berlaku selepas 20 jam 10 minit pada suhu 27 sehingga 28 °C. Purata panjang larva yang baru menetas ialah 1.86 mm, dan purata tinggi ialah 0.460 mm. Pundi-pundi kuning telur dan titisan minyak masing-masing habis digunakan 63 dan 65 jam selepas menetas. Larva yang berumur 1 hari mempunyai purata panjang 2.253 mm, dan purata tinggi 0.615 mm. Pada umur 49 hari, purata panjang dan tinggi masing-masing meningkat sehingga 27.153 mm dan 9.190 mm.

Spin larva mula kelihatan pada umur 7 sehingga 10 hari. Spin dorsal mencapai panjang maksimum pada umur 28 sehingga 30 hari dengan purata

panjang adalah 5668.30 μm manakala spin ventral telah mencapai purata panjang maksimum ialah 4415.44 μm pada umur 25 sehingga 28 hari. Seterusnya, spin mulai memendek dan berubah menjadi tulang sirip keras.

Pigmen mata mulai kelihatan pada hari ke 3 sehingga hari ke 4 selepas menetas. Mulut larva mulai terbuka pada hari ke 2 atau 45 jam selepas menetas. Hubungan diameter mata dengan bukaan mulut adalah kuadratik dengan persamaan $MG = -0.2E\text{-}05Ed^2 + 0.3992Ed + 215.05$ dan $R^2 = 0.98$. Bintik-bintik hitam telah kelihatan pada pangkal ekor, sirip dorsal dan sirip anal apabila larva mencapai umur 25 hingga 30 hari. Jumlah bintik-bintik ini telah bertambah dan tersebar ke arah kepala dan lengkap pada umur 40 hingga 45 hari. Pada masa itu, ia kelihatan serupa ikan juvenil.

Gigi larva ikan kerapu tikus mulai kelihatan pada umur 15 hari, tetapi jumlah bilangan gigi masih sedikit dan pendek. Tinggi dan lebar gigi meningkat dengan pertambahan umur. Pada umur 30 hari, gigi larva semakin rapat dengan purata tinggi dan lebar masing-masing adalah 110.18 μm dan 40.13 μm.

Dari aspek histologi, tidak terdapat lipatan di dalam saluran penghadaman larva ikan sebelum berumur 5 hari. Pembentukkan perut mulai kelihatan pada umur 30 hari. Perut larva mulai lengkap pada umur 45 hari. Hati larva mulai kelihatan pada umur 5 hari, manakala pankreas mulai kelihatan pada umur 10 hari. Akiviti enzim protease meningkat dengan pertambahan umur.

Makanan mula ditemui di dalam saluran penghadaman pada umur 3 hari atau 66 jam selepas menetas. Purata jumlah makanan yang ditemui di dalam saluran penghadaman adalah 2 ind./larva. Rotifer yang mula-mula dijumpai di dalam saluran penghadaman mempunyai saiz purata 55.44 – 96 μm. Copepoda dengan saiz purata 104.16 – 116.28 μm mula pertama kali dimakan pada umur 6 hari, manakala nauplii *Artemia* pada umur 15 hari.

Larva yang berumur 1 hari mempunyai berat 0.067 mg dan berat akan meningkat sehingga 141.114 mg pada umur 49 hari dengan purata kadar tumbesaran 2.88 mg/hari. Kadar kemandirian larva sehingga 49 hari adalah 3.03%.

Larva yang diberi makan dengan *Artemia* memperolehi kadar tumbesaran 1.91 mg/hari menunjukan perbezaan yang signifikan ($p < 0.01$) berbanding daripada larva yang diberi makanan buatan mulai pada umur 35, 40 dan 45 hari. Kadar tumbesaran larva yang diberi makanan buatan pada umur 35, 40 dan 45 hari masing-masing adalah 0.09 mg/hari, 0.022 mg/hari and –0.02 mg/hari. Larva yang diberi makanan buatan paling awal mempunyai tumbesaran lebih baik berbanding larva yang lambat diberi makan buatan. Larva yang diberi makanan buatan pada umur 35 hari adalah mempunyai kadar tumbesaran lebih tinggi daripada larva yang diberi makanan buatan pada umur 40 hari. Purata kadar kemandirian larva yang diberi makan *Artemia* adalah 21.34%, manakala larva yang diberi makanan buatan pada umur 35, 40 dan 45 hari masing-masing adalah 8, 18 dan 19.33%.

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I certify that an Examination Committee met on 1 February 2002 to conduct the final examination of Usman Bulanin on his Doctor of Philosophy thesis entitled "Study on the Development of Embryo and Larva of the Humpback Grouper (*Cromileptes altivelis* Valencee) and Its Feeding Behaviour" in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulations 1981. The committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follows:

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AF	= After fertilization
ANOVA	= Analysis of variance
C	= Control
cm	= Centimetre
CRD	= Complete randomized design
D	= Day
DAH	= Day after hatching
DO	= Dissolved oxygen
Dsp	= Dorsal spine
E	= *Epinephelus*
Ed	= Eye diameter
F	= Fish
g	= Gram
GR	= Growth rate
GRH	= Growth rate height
GRL	= Growth rate length
GRW	= Growth rate weight
H	= Height
h	= Hours
HAH	= Hours after hatching
HE	= Hematoxylin eosin
HUFA	= High unsaturated fatty acid

Ind.	= Individual
U	= Unit
IUC	= Inter university center
L	= Length
L	= Litre
L	= *Lutjanus*
m	= Metre
MG	= Mouth gape
mg	= Milligram
mm	= Millimetre
ml	= Millilitre
µm	= Micron metre
N	= Nauplii
Od	= Oil droplet
ppm	= Parts per million
ppt.	= Parts per thousand
r	= Radius
SR	= Survival rate
V	= Volume
Vsp.	= Ventral spines
W	= Weight
Ys	= Yolk sac