REPRODUCTIVE BIOLOGY AND LARVAL REARING OF BLUE SWIMMING CRAB, *PORTUNUS PELAGICUS* (LINNAEUS, 1758)

By

E F R I Z A L

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia in Fulfilment of the Requirement for the Degree of Doctor of Philosophy

August 2006

DEDICATION

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To my parents,

J. Halawa & Maizarni,

my brothers and sister, Hendrizal, Amirwan, Srina Marinawati & Linda Riani
thank you for your love, understanding and support.

*****

To my beloved wife and child

Dedek Hidayati, and M.Teguh Dhjya Ulhaq,

Your love, encourragment and patience sustained me through, thank you.

and

To my in laws,

Thank you for your patience and understanding.

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

REPRODUCTIVE BIOLOGY AND LARVAL REARING OF BLUE SWIMMING CRAB, *PORTUNUS PELAGICUS* (LINNAEUS, 1758)

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Chairman : Associate Professor Aziz Arshad, PhD

Faculty : Science

Portunus pelagicus broodstock caught from the coastal region of Port Dickson, Negeri Sembilan, Malaysia in August 2004, and conditioned in the Hatchery Unit of Marine Research Station, UPM were used for the reproductive experiments including larval rearing studies. Various aspects of the reproductive biology of blue swimming crab, *P. pelagicus* were studied. Generally, sexual dimorphism and reproductive system of male and female blue swimming crab were observed to be similar to most other decapods crustaceans. The pubertal molt, the abdominal and gonopores structures of the female showed changes that are generally accepted as external morphological indications of sexual maturity. Unlike the female, the male showed pre-pubertal (loss of the attachment of the abdominal flap to the cephalothorax) rather than pubertal molt. The ovaries and testes were classified into five and three development stages respectively, and the ovarian histology of each stage was characterized. The ovarian stages correlated closely with the gonadosomatic index (GSI), the characteristics

of ovarian histology and oviposition period. Fecundity estimates ranged from 148,897 to 835,401 eggs with brood size highly correlated to carapace width (CW), carapace length (CL), body weight (BW) and egg batch weight (EBW). The complete embryonic development of the blue swimming crab, *P. pelagicus* was described based on morphological features observed in live eggs. Periods of development were defined in sequence of 12 to 24 hours each and in relation to the time of embryonic development. Eight periods were described and illustrated for *P. pelagicus*. Embryonic development from recently spawned eggs (egg-mass appeared yellow and compact) to hatching lasted 8 days at 28-30^{o}C. The larval stages included four zoeal stages and one megalopa. The megalopa molted to the first crab instar. The zoeae and megalopa were very similar to those of other portunids. The first and the second zoeal stages spanned 3-4 days each, the third and fourth stages were 2-3 days each, and the megalopa was 3-4 days. The first crab instar emerged 15- 18 days after hatching.

The egg incubation period decreased exponentially from 8.33 to 6.67 days with the increase in temperature in the range 28-34^{o}C. The best fertilization and hatching rates of eggs were obtained at temperature in the range 28-30^{o}C and 28-32^{o}C, respectively. Relationships between temperature, and incubation period, fertilization and hatching rate of eggs were found to be quadratic.

The development duration and survival rate of the blue swimming crab larvae fed with tested diets were significantly higher compared to the control. Thirty percent of first zoea successfully developed to the first crab stage when fed with the *Artemia* nauplii alone whereas, 15.56% molted from the first zoea to megalopa when fed with the combination diets containing *Nannochloropsis oculata* + *Artemia* nauplii (15.56%) and *N. oculata* + rotifers *Branchionus plicatilis* + *Artemia* nauplii (5.56%). The molting of the first zoea to third zoea (22.67%) was achieved through the single use of rotifers and larval development from first zoea to second zoea (10.00%) was achieved by using *N. oculata* + rotifers *B. plicatilis*. No development beyond pre-metamorphic first zoea was observed when the larvae were fed solely with *N. oculata* diet but the results showed they survived longer compared to the control.

A continuous fluorescent light of 3000-3500 lux supported the highest mean survival (27.78%) from hatching to successful metamorphosis of first crab instar. The first zoeal stages reared under 1000-1500 lux did not reach the third zoeal stages. All larvae died after 14 days.

The influence of temperature was more evident on the survival rate and larval development duration during the first crab stages (C1) ($P<0.05$). The highest and lowest larval survival rates until C_1 stages were at 30^oC (36.67%) and 34^oC (12.22%) respectively. The second best survival rate of C1 (31.11%) was obtained at 28^oC. The shortest larval development (3.67-

4.00 days) occurred at 30-34^{o}C. At 26^{o}C and 28^{o}C larval development of C1 stage took around 5.67 days (P>0.05).

In order to achieve optimal growth and survival in larviculture, blue swimming crab larvae is recommended to be reared at salinities in the range of 28-30 ppt. The development duration of C_1 was relatively shorter (3.33-4.00 days) at all salinity levels. The relationships between salinity, and successful metamorphosis and development duration of C_1 were quadratic.

A high stocking density (40-60 larvae/L) consistently produced low successful metamorphic development throughout the experimental period resulting in only 19.11-20.00% final survivals. The low stocking density levels (20-30 larvae/L) tested in this study resulted in only 22.67-23.11%, which were not significantly different (P>0.05) among the five treatments. The relationships between stocking density, and larval development duration and successful metamorphosis of C_1 were linear and quadratic respectively.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

BIOLOGI PEMBIAKAN DAN PEMELIHARAAN LARVA KETAM RENJUNG, *PORTUNUS PELAGICUS* (LINNAEUS, 1758)

Oleh

EFRIZAL

Ogos 2006

Pengerusi : Profesor Madya Aziz Arshad, PhD

Fakulti : Sains

Induk ketam renjung, *Portunus pelagicus* yang dikumpulkan dari kawasan perairan Port Dickson, Negeri Sembilan, sekitar Ogos 2004 dan disesuaikan di Unit Penetasan Stesen Penyelidikan Marin Universiti Putra Malaysia, telah digunakan untuk eksperimen pemeliharaan larva. Pelbagai aspek pembiakan biologi ketam renjung, *P. pelagius* telah dikaji. Seks dimorfisme dan sistem pembiakan jantina am ketam renjung didapati serupa dengan kebanyakan dekapod krustasia lain. Persalinan kulit mencapai kedewasaan, struktur abdomen dan liang gonad betina menunjukkan perubahan yang kebiasaannya diterima sebagai tanda morfologi luaran bagi kematangan seksual. Berbeza dari ketam betina, ketam jantan menunjukkan persalinan kulit pra-kedewasaan (kehilangan pautan bahagian abdomen sefalotoraks) dan tidak bersalin kulit mencapai kedewasaan. Bahagian ovari dan testis telah diklasifikasikan kepada lima dan tiga peringkat perkembangan, serta histologi ovari pada setiap peringkat telah masing-masing dikategorikan. Peringkat ovari berkorelasi rapat dengan indeks gonadosomatik fekunditi

telur (GSI), sifat histologi ovari dan jangkamasa pengaruhan telur. Julat jangkaan fekunditi telur dari 148,897 sehingga 835,401 adalah berkorelasi tinggi dengan kelebaran karapas (CW), panjang karapas (CL), berat badan (BW) dan gumpalan berat telur (EBW). Perkembangan embrio lengkap ketam renjung *P. pelagicus* dibincangkan berdasarkan kepada ciri-ciri morfologi yang dilihat pada telur sebenar. Jangkamasa perkembangan dikenalpasti dalam masa 12 hingga 24 jam dan ianya berkait dengan jangkamasa perkembangan embrio. Sebanyak lapan jangkamasa perkembangan telah dibincangkan dan diilustrasikan bagi *P. pelagicus*. Perkembangan embrio dari telur yang baru dipersenyawakan (jisim telur kelihatan kekuningan dan padat) sehingga kepada proses penetasan berlangsung selama 8 hari pada suhu 28-30^{o}C. Peringkat larva terdiri dari empat peringkat zoea dan satu peringkat megalopa. Megalopa berekdisis dan bertukar kepada ketam instar pertama. Zoea dan megalopa hampir serupa dengan ketam portunids yang lain. Tempoh masa bagi setiap dua peringkat pertama zoea adalah selama 3-4 hari, dua peringkat seterusnya mengambil masa selama 2-3 hari dan megalopa pula mengambil masa selama 3-4 hari untuk mencapai peringkat pertama ketam yang didapati selepas 15-18 hari.

Tempoh masa bagi telur dalam pengeraman menurun secara eksponen dari 8.33 sehingga 6.67 hari dengan peningkatan suhu dalam julat 28-34^{o}C. Kadar persenyawaan dan penetasan telur yang terbaik boleh didapati pada

julat suhu 28-30^{o}C dan 28-32^{o}C. Hubungan antara suhu dan jangka masa pengeraman dan kadar penetasan telur didapati kuadratik.

Kadar perkembangan dan kemandirian bagi larva ketam renjung yang diuji menggunakan ujian gizi didapati lebih tinggi bererti berbanding dengan kawalan. Tiga puluh peratus larva zoea awal berjaya berkembang ke peringkat ketam pertama dengan menggunakan gizi *Artemia* nauplii sahaja sebagai sumber pemakanan. Sebanyak 15.56% berekdisis daripada zoea awal kepada megalopa dengan pemakanan yang menggunakan kombinasi gizi yang mengandungi *Nannochloropsis oculata* + *Artemia* nauplii (15.56%) dan *N. oculata* + rotifers *Branchionus plicatilis* + *Artemia* nauplii (5.56%). Ekdisis zoea pertama sehingga zoea ketiga (22.67%) telah dicapai melalui penggunaan diet rotifers *B. plicatilis* sahaja dan perkembangan larva dari peringkat pertama zoea sehingga ke peringkat kedua zoea (10.00%) dicapai menggunakan *N. oculata* + diet rotifers *B. plicatilis*. Tiada perkembangan melepasi peringkat pra-metamorfik zoea pertama dapat diperhatikan apabila larva diberi gizi *N. occulata* tetapi keputusan menunjukkan tahap kemandiriannya adalah lebih lama berbanding kawalan.

Pencahayaan yang berterusan pada kadar 3000-3500 lux menyokong min kemandirian yang tertinggi (27.78%) dari peringkat penetasan kepada metamorfosis lengkap bagi ketam instar pertama. Pemeliharaan peringkat

pertama zoea dibawah 1000-1500 lux tidak menghasilkan peringkat ketiga zoea; kesemua larva mati selepas 14 hari.

Pengaruh suhu lebih ketara pada kadar kemandirian dan tempoh perkembangan larva di peringkat pertama ketam (C_1) ($P<0.05$). Kadar kemandirian larva yang tertinggi dan terendah sehingga peringkat C_1 telah dirangsang pada suhu 30^oC (36.67%) dan 34^oC (12.22%). Kadar kemandirian kedua terbaik bagi C_1 (31.11%) didapati pada suhu 28^oC. Perkembangan larva terendah (3.67-4.00 hari) berada pada suhu 30-34^oC. Pada suhu 26^oC sehingga 28^oC, perkembangan larva bagi peringkat C_1 mengambil masa lingkungan 5.67 hari ($P>0.05$).

Untuk mencapai pertumbuhan dan kemandirian optimum dalam aktiviti kultur semasa larva iaitu sebelum pelepasan ke kolam, larva ketam renjung boleh dipelihara pada tahap julat kemasinan dalam julat 28-30 ppt. Jangkamasa perkembangan C_1 lebih singkat (3.33-4.00 hari) pada kesemua paras kemasinan dan tidak bererti secara statistic ($P>0.05$). Hubungan antara kemasinan dan perlengkapan metamorfosis dan tempoh masa perkembangan C_1 adalah bersifat kuadratik.

Sepanjang tempoh eksperimen, pelepasan dengan kepadatan tinggi (40-60 larva/L) secara berterusan akan menghasilkan kejayaan perkembangan metamorfik yang rendah. Di akhir perkembangan, kejayaan kemandirian metamorfik yang dihasilkan adalah sebanyak 19.11 hingga 20.00% pada

kepadatan tinggi dan 22.67% hingga 23.11% pada kadar pelepasan kepadatan rendah (20-30 larva/L). Secara statistik tiada perbezaan yang bererti ($P>0.05$) antara lima rawatan ini. Keputusan bagi analisis regresi polinominal menunjukkan hubungan antara kepadatan dan perlengkapan metamorfosis dan tempoh masa perkembangan C_1 adalah bersifat kuadratik dan linear masing-masing.

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I certify that an Examination Committee has met on 18th August 2006 to conduct the final examination of Efrizal on his Doctor of Philosophy thesis entitled "Reproductive Biology and Larval Rearing of Blue Swimming Crab, *Portunus pelagicus* (Linnaeus, 1758)" 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 follow:

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This thesis submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Doctor of Philosophy. The members of the Supervisory Committee are as follows:

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Date: 14 DECEMBER 2006

DECLARATION

I hereby declare that thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.

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EFRIZAL

Date: 19 NOVEMBER 2006

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