

MORPHOMETRIC VARIABILITIES IN THE ADULT
POPULATIONS OF MANGROVE HORSESHOE
CRAB *CARCINOSCORPIUS ROTUNDICAUDA*
(LATREILLE), EMBRYONIC DEVELOPMENT AND
THEIR LARVAL BEHAVIOUR UNDER DIFFERENT
CULTURE CONDITIONS

SRIJAYA T.C.

DOCTOR OF PHILOSOPHY
UNIVERSITI MALAYSIA TERENGGANU
MALAYSIA

2011

**MORPHOMETRIC VARIABILITIES IN THE ADULT
POPULATIONS OF MANGROVE HORSESHOE
CRAB *CARCINOSCORPIUS ROTUNDICAUDA*
(LATREILLE), EMBRYONIC DEVELOPMENT AND
THEIR LARVAL BEHAVIOUR UNDER DIFFERENT
CULTURE CONDITIONS**

SRIJAYA T.C.

**Thesis submitted in Fulfillment of the requirement for
the Degree of Doctor of Philosophy in the
Institute of Tropical Aquaculture
Universiti Malaysia Terengganu**

May' 2011

Dedicated to

All well wishers.....

And

An ancient living wonder - Horseshoe crab

Abstract of thesis presented to the Senate of Universiti Malaysia
Terengganu in fulfillment of the requirement for the
Degree of Doctor of Philosophy

**MORPHOMETRIC VARIABILITIES IN THE ADULT POPULATIONS OF
MANGROVE HORSESHOE CRAB *CARCINOSCORPIUS
ROTUNDICAUDA* (LATREILLE), EMBRYONIC DEVELOPMENT AND
THEIR LARVAL BEHAVIOR UNDER DIFFERENT CULTURE
CONDITIONS**

SRIJAYA T.C.

MAY' 2011

Chairman : Associate Professor Anuar bin Hassan, Ph.D.
Member : Anil Chatterji, Ph.D.
Professor Faizah Shaharom, Ph.D.
Institute : Institute of Tropical Aquaculture

Carcinoscorpius rotundicauda (Latreille) is one of the four extant species of horseshoe crab abundantly found in the mangrove ecosystem in Peninsular Malaysia. The morphometric variability in two populations collected from Setiu (Terengganu) and Gelang Patah (Johor) was studied by applying allometric analysis which was fitted well in describing the morphometric diversity between the adult two different populations. The carapace length and width, and telson length were; 2.65, 1.26 and 16.26% higher in males of Setiu population as compared to male specimens collected from Gelang Patah (Johor) whereas 9.46, 7.90 and 13.34%, respectively in female population of Setiu as compared to female population of Gelang Patah (Johor). The growth in all body parts of both populations followed an

isometric growth pattern showing exponential constants values ~ 1 in all relationships. Similarly the body weight of the specimens collected from Setiu was greater (males= 145 ± 18.06 g; females= 250 ± 13.79 g) than in Gelang Patah population (males= 126 ± 18.25 g; females= 170 ± 21.79 g). In males of Setiu population, a maximum growth in weight was recorded ($b=2.12$). In males of Gelang Patah population, maximum regression coefficient values were recorded ($b=2.93$) showing that the increment in body weight was greater than increment in carapace width confirming a significant relationship. The regression coefficients (b -value) ranged from 0.41 (females of Setiu) to 2.93 (males of Gelang Patah) and length-weight relationship did not follow isometric growth except the total length and body weight relationship of males of Gelang Patah population. The trend of energy utilization from the endogenous reserves by the developing embryos of the horseshoe crab from 0 to 40 days of incubation was studied in detail. The dry weight, insoluble proteins, carbohydrates, glycogen and lipid values were decreased from 0 to 40 days whereas wet weight, water content, ash content and soluble proteins increased considerably with the development of the eggs. Microelements such as Cu^{++} , Fe^{++} , Zn^{++} and Cr^{++} also showed maximum variations during 1st and 2nd moulting stages of the eggs. Maximum amount of lipids (71.2%) carbohydrate (39.4%), insoluble proteins (24.9%), glycogen (22.3%) and microelements (Cu^{++} , Fe^{++} , Zn^{++} and Cr^{++}) were required during 10-35 days of development of the embryos.

Variation in oxygen consumption by the trilobite larvae of this species was not much at temperatures 10 and 20° C and the lowest oxygen consumption

was 0.36 mg/ml at 30° C and the highest (1.98 mg/ml) at 40° C during the first 4 hours of the experiment. The oxygen consumption rate was maximum (0.06 g/hr) at a temperature 40° C whereas minimum (0.03 g/hr) at a temperature 10° C ($P < 0.05$; $r = 0.99$). The oxygen consumption was recorded maximum between pH 7 and 9 during the first 4 hours with a sudden increase at 48 hours at pH 4. The oxygen consumption rate was higher (0.08 g/hr) under light conditions as compared to dark conditions (0.05 g/hr) ($r=0.88$); however, the rate of oxygen consumption did not show much variation in different salinities.

The behavioral changes of trilobite larvae with regard to different colour and lights showed a maximum congregation in white coloured zone ($n=12$) followed by yellow ($n=9$), orange ($n=8$) and then blue ($n=7$) colour zones. The Morisita's index values were always below 1 (0.98) which further confirmed preference of trilobite larvae towards the white colour. Similarly, the trilobite larvae showed more preference towards infrared (45% congregation) than ultra-violet (33%) lights when the positions of the light sources were kept horizontal and vertical respectively.

The congregational behavior of the trilobite larvae with respect to factors like tide time, tidal phases and moon positions was also studied in detail by applying cluster analysis and Multidimensional Scaling (MDS) using PRIMER v.6 software. The pattern of congregation of the larvae showed 12 associations where the similarity between these associations for both dark and light conditions was same at 95% level. A maximum congregation of the

larvae was occurred between 270 and 360° sector towards the northern pole of the experimental tank in coincidence with the new moon phase (absolute magnitude of moon ~42 AU). When the absolute magnitude of the moon was ~32 AU, a clear shift of the congregation from northern to western pole of the tank (180-270°) was observed during full moon phase ($P < 0.05$). A maximum congregation was observed towards the western pole (absolute magnitude of the moon ~32 AU) and towards the northern pole (absolute magnitude of the moon ~42 AU). The congregation of the trilobite larvae was highly influenced by the lunar phases and tidal amplitudes irrespective of the time and photoperiod.

The most suitable environmental parameters like temperature and salinity for rearing of the trilobite larvae under controlled condition was also identified. The survival of the larvae was only up to the 4th month at a temperature of 10° C with a maximum weight (79.6 mg) at salinity 20 ppt. A maximum 443 mg weight was recorded during 12 months when the temperature and salinity were 20° C and 40 ppt respectively. At 20° C temperature and salinities 20 and 30 ppt, all the larvae were found dead after 9th month. Specific growth rate (SGR), relative growth rate (RGR) and growth increments at 20° C were the highest at 40 ppt salinity during 8th month immediately after 4th moulting stage of the larvae. The maximum attainable weight (W_{∞}) was 900 mg in this group. A maximum weight (836.4 mg) was recorded at temperature of 30° C when the salinity was 40 ppt. The maximum survival of the larvae was 40% at 30° C temperature when the salinity was 30 ppt ($p < 0.05$). The survival rate of the larvae was relatively low

(14%) at temperature 30° C and salinity 40 ppt although a maximum attainable weight 4750 mg was observed. All larvae were found dead within a month of the culture period at temperature 40° C in all salinity ranges. The results of the present study showed that for rearing the larvae of *C. rotundicauda*, a combination of 30° C temperature and 30 ppt salinity are the ideal environmental parameters.

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

KEPELBAGAIAN MORFOMETRIK DALAM POPULASI DEWASA BELANGKAS PAYA BAKAU *CARCINOSCORPIUS ROTUNDICAUDA* (LATREILLE), PERKEMBANGAN EMBRIO DAN KELAKUAN LARVA DIBAWAH KULTUR PELBAGAI KEADAAN

SRIJAYA T.C.

MAY' 2011

Pengerusi : Professor Madya Anuar bin Hassan, Ph.D.

Ahli : Anil Chatterji, Ph.D.

Professor Faizah Shaharom, Ph.D.

Institut : Institut Akuakultur Tropika

Belangkas, *Carcinoscorpius rotundicauda* merupakan salah satu daripada spesies belangkas yang ditemui di paya bakau Semenanjung Malaysia. Kesesuaian morfometrik antara populasi Setiu (Terengganu) dan Gelang Patah, Johor dikaji menggunakan analisis morfometrik bagi menerangkan kepelbagaian antara dua populasi. Panjang dan lebar carapace, dan juga panjang telson ialah 2.65, 1.26 dan 16.26% lebih tinggi untuk populasi jantan Setiu (Terengganu) dibandingkan dengan specimen jantan dari Gelang Patah (Johor) manakala, 9.46, 7.90 dan 13.34% bagi populasi betina dari setiu dibandingkan dengan populasi betina dari Gelang Patah, Johor. Kedua-dua populasi mengikut tumbesaran isometrik bereksponansi tetap~1 dalam semua aspek. Berat badan specimen dari Setiu (jantan= 145 ± 18.06 g; betina= 250 ± 13.79 g) berbanding populasi Gelang Patah (jantan= 126 ± 18.25 g; betina= 170 ± 21.79 g). Bagi populasi jantan Setiu, pertambahan berat

maxima dicatat, $b=2.12$ manakala populasi jantan Gelang Patah regresi maksima dicatat ($b=2.93$) yang menunjukkan berat badan bertambah lebih besar dari lebar badan dengan hubungkait signifikan. Pekali regresi (nilai- b) bernilai antara 0.41 (betina Setiu) sehingga 2.93 (jantan Gelang Patah) dan hubung kaitan panjang-lebar tidak mengikut ketumbuhan isometric kecuali panjang mutlak dan berat badan populasi jantan Gelang Patah.

Penggunaan tenaga daripada sumber dalaman embrio belangkas dari hari 0 hingga 40 dikaji teliti. Berat kering, protein tak larut, karbohidrat, glikogen dan lipid berkurang nilainya dari hari 0 hingga 40 manakala berat basah, air, abu dan protein larut meningkat dengan perkembangan telur. Elemen mikro seperti Cu, Fe, Zn dan Cr menunjukkan variasi maksimum selama 1 dan 2. Lipid (71.2%), karbohidrat (39.4%), protein tak larut (24.9%), glikogen (22.3%) dan elemen mikro (Cu, Fe, Zn dan Cr) diperlukan semasa hari ke 10-35 perkembangan embryo.

Variasi penggunaan oksigen oleh larva trilobat species ini kurang untuk suhu 10 dan 20 °C dan penggunaan oksigen terendah ialah 0.36 mg/ml untuk 30 °C dan paling tinggi 1.98mg/ml untuk suhu 40 °C manakala paling minima 0.03g/j pada suhu 10 °C ($p<0.05$; $r=0.99$). Penggunaan oksigen dicatat maksima antara pH 7 dan 9 pada 4 jam pertama dan meningkat pada jam ke 48 pada pH 4. Kadar penggunaan oksigen lebih tinggi (0.08 g/j) dibawah cahaya berbanding gelap (0.05g/j) ($r=0.88$) walhal, kadar guna oksigen tidak menunjukkan variasi dalam saliniti berbeza.

Laman perilaku larva "trilobite" berkaitan dengan warna yang berbeza dan lampumenunjukkan jemaat maksimum di zon berwarna putih (n=12) diikuti warna kuning (n=9), oren (n=8) dan biru (n=7). Nilai Indeks Morisita sentiasa bawah 1 (0.98) yang menunjukkan keutamaan larva "trilobite" terhadap warna putih. Sehubungan dengan itu, larva "trilobite" menunjukkan keutamaan terhadap inframerah (45% perkumpulan) berbanding dengan ultraungu (33%) apabila posisi sumber cahaya disusun mendatar dan menegak.

Kelakuan perkumpulan maksima oleh larva "trilobite" disebabkan oleh keadaan dan waktu pasang surut dan juga posisi bulan telah dikaji menggunakan analisis Multidimensional Scaling (MDS) software PRIMER v.6. Corak Perkumpulan larva menunjukkan 12 tabiat dan keseragaman antara tabiat dalam keadaan gelap dan cahaya adalah 95%. Perkumpulan maksima larva berlaku antara sudut 270 dan 360° menghadap kutub utara tangki experiment berikutan dengan fasa bulan (magnitud bulan ~42AU). Apabila magnitud bulan menjadi ~32 AU, perubahan perkumpulan dari Utara kepada kutub Barat tangki (180-270° berlaku semasa bulan penuh (P<0.05) Perkumpulan maksima berlaku terhadap kutub Barat (magnitud bulan ~32AU) dan terhadap kutub utara (magnitud bulan ~42 AU). Perkumpulan larva "trilobite" dipengaruhi fasa bulan dan amplitude pasang surut tanpa pengaruh waktu dan fasa cahaya.

Parameter persekitaran yang paling sesuai seperti suhu dan saliniti bagi penyelenggaraan larva trilobite di bawah keadaan terkawal juga dikenalpasti

Kelangsungan hidup larva hanya sampai 4 bulan pada suhu 10 °C dengan berat maksima 79.6 mg pada salinity 20ppt. Berat maksimum 443mg tercatat selama 12 bulan pada suhu dan salinitas yang 20° C dan 40 ppt masing-masing. Pada suhu 20° C dan salinity 20 dan 30 ppt, semua larva mati selepas 9 bulan. Kadar Pertumbuhan Spesifik (SGR), Kadar pertumbuhan Relatif (RGR) dan tumbesaran pada suhu 20° C adalah paling tinggi pada salinitas 40 dalam bulan ke 8 seurus selepas fasa penyahkulitan ke-4. Bagi kumpulan ini, berat (W_{∞}) adalah 900mg. Berat maksima (836.4 mg) tercatat pada suhu 30° C, salinitas 30 ppt ($p < 0.05$). Kadar hidup larva adalah rendah (14%) pada suhu 30° C dan salinitas 40 ppt walaupun berat mencapai 4750mg. larva Semua ditemui mati dalam masa satu bulan tempoh kultur pada suhu 40° C disemua julat salinitas. Keputusan kajian ini menunjukkan bahawa untuk penyelenggaraan larva *C.rotundicauda*, kombinasi suhu 30° C dan salinity 30 ppt adalah parameter persekitaran yang ideal.