Temporal Variations in the Development, Growth, and Survival of the Larvae of Queen Conch *Strombus gigas* under Experimental Cultures

Variación Temporal en el Desarrollo, Crecimiento y Sobrevivencia de las Larvas del Caracol Rosado *Strombus gigas* Provenientes de Cultivos Experimentales

Variations Temporelles du Développement, de la Croissance et de la Survie des Larves de *Strombus gigas* dans des Cultures Expérimentales

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ABSTRACT

Larvae of *Strombus gigas* from egg masses of natural ovoposition were reared under laboratory conditions for one year during the reproductive season (from March to September) in Chinchorro Bank. The same types of rearing containers and techniques were employed. Egg masses are incubated for 3-5 days in 25-L containers, and cultured in 4-L containers. Larvae were fed with *Tetraselmis suecica* at a concentration of 1,000 algal cells/ml, density of 200 larvae/L, temperature of 29º ± 1º C, and a natural photoperiod. At hatching, the larvae from March, April, May, and September cultures presented 2 velar lobes, 1.5 shell whorls, and right tentacle (1-3 days), whilst the June, July and August, developed 4 velar lobes, 2.0 shell whorls, and right tentacle (1-3 days). The mean shell length at hatching was 300 ± 1.52 µm for all culture months. The development of proboscis began between 11 to 16 days, for June to August cultures and in 19-21 days for March, April, May, and September cultures. Settlement was first observed at 27 days in larvae from June to August cultures (42%); for March, April, May, and September cultures it was in 29 days in 29% of larvae. The maximum shell lengths have been obtained in August and September (908 µm both), whilst the minimum was registered for April with 867 µm. The growth rate varied from 22.33 µm/day to 30.00 µm/day. With regard to survival, the percentages obtained in June and July (38%) were highest in relation to September values (20%). For the other months, survival was 22% for March culture, 34% in April, 37% for May, and 28% in August culture. The results obtained in this work, demonstrate that the development, growth, and survival of the larvae from the intermediate months (June to August) of the reproductive season were much better ($F = 4.01$, $p < 0.05$, Tukey < 0.01), than in early and later months (March, April, May, and September), which can be related to the parents maturity.

KEYWORDS: *Strombus gigas*, temporal variations, development, growth, survival

INTRODUCTION

The queen conch *Strombus gigas* (Linné 1758), is distributed throughout the shallow waters of the Caribbean Islands, Central America, north-eastern South America to Brazil, Florida, the Bahamas, and Bermuda (Warmke and Abbott 1961, in Creswell 1994). Conch resources throughout the region have been subject to heavy fishing pressure, which has resulted in precipitous declines in natural populations (Creswell 1994).

The critical status of *S. gigas* populations in many localities around the Caribbean has been a topic of concern and the subject of many studies during the last 30 years. Overfishing and lack of fisheries management have been cited as the major causes in the decline of natural conch populations (Weil and Laughlin 1994).

In recent years, there has been increasing interest in stock rehabilitation through release of mass production of hatchery -reared juveniles (Berg 1976, Brownell 1977, Brownell and Stevely 1981, Weil and Laughlin 1982, Appeldoorn and Ballantine 1983, Siddall 1984, Davis et al. 1987, Davis et al. 1993, Davis 2000 a,b). Eggs (Robertson 1959) and larval development (D'Asaro 1965, Davis 1994a) of *S. gigas* have been described for over 25 years.

Mariculture has the potential to supplement fisheries landings and contribute to stock rehabilitation, but considerable constraints to large-scale production still exist (Appeldoorn 1994).

Better assessments of management have been realized (Brownell and Stevely 1981, Goodwin 1981, 1983), food quality studies of larval and juveniles have been conducted (Pillsbury 1985), aspects of physiology and larval development has been investigated (Sanders 1984, Laughlin and Weil 1985, Davis et al. 1993, Davis 1994a, 2000 a,b), metamorphosis inducers has been tested, with promising results (Sidall 1983, Davis et al. 1990, Davis 1994 a,b, 2000 a,b). Appeldoorn (1994) suggested that the greatest needs be in post-larval growout, particularly the development of a cost-effective artificial feed and the reduction of predation (e.g. identifying optimum habitats, pre-adapting juveniles to field conditions, predator control).

For another hand, recruitment is a key area requiring further research. Knowledge of the production of larvae is important for understanding stock structure and the variability in stock abundance due to natural short-term recruitment fluctuations. The study of larval recruitment can be determining the factors affecting larval production.

In this sense, and since that the literatures on the role of temporal variations on recruitment in experimental cultures are poorly know, the goal of the present work was study these in experimental cultures on development, growth, and survival of larvae of *S. gigas* to evaluate their impact on the larval recruitment.

MATERIALS AND METHODS

These experimental cultures were designed to test temporal variations in development, growth, and survival to settlement for veligers of *S. gigas*.

The study was conducted from March to September at the Marine Biology Laboratory in CINVESTAV. Newly laid egg masses were collected from a spawning population in Alacranes Reef during the reproduction season. For the experiment, three fertilized egg masses were taken each month, from under a spawning female conch to ensure species identity and to help estimate hatch date (Davis et al. 1987), from a depth of 4 m.

These were transported individually in a closed thermic container with seawater to the laboratory, where sand particles were removed. Cleaned eggs were placed over a 300 µm mesh and kept immersed in a 25-liter tank with seawater filtered through 2 µm cotton filters and UV sterilized at ambient temperature (29 ± 1°C). All experimental cultures were not run simultaneously. Each hatch of veligers was culture for one month under conditions mentioned above.

A sample of 30 newly hatched veligers was measured to determine initial size. A dissecting microscope equipped with an ocular micrometer was used to measure shell length from apex to siphonal canal at 10x magnification.

For each egg mass, three experimental cultures were set up in 4-L containers with a density of 200 larvae/L. Larvae were fed daily the algae *Tetraselmis suecica* at a concentration of 1000 cells/ml. The experiments were run at 29 ± 1ºC, and the seawater was renewed totally every 24 hours. Each morning, veligers were transferred to new containers with fresh seawater filtered through 10 and 15 µm cotton filters. The veligers and treatment water were removed by pouring them through a submerged sieve with the appropriate size mesh (180 - 300 µm). A wash bottle filled with water was used to move the veligers from the sieve into the new container.

Daily, 30 larvae were removed at random from each replicate for developmental observations. To avoid damaging veligers during observations, they were removed carefully with a pipet and placed in a seawater-filled Petri dish.

Based on larval development nine developmental stages were identified and recorded:

i) hatching;
ii) two velar lobes and 1.5 - 2.0 whorls;
iii) four velar lobes, 2.0 - 3.5 whorls without heart (4a);
iv) four velar lobes, 2.0 - 3.5 whorls and adult heart (4b);
v) six velar lobes, 3.0 - 4.0 whorls without proboscis (6a);
vi) six velar lobes, 3.0 - 4.0 whorls with proboscis (6b);
vii) six velar lobes, 4.0 - 4.5 whorls without radula (6c);
viii) six velar lobes, 4.0 - 4.5 whorls with radula (6d); and
ix) Settlement (6e)

Developmental stages at a given age were based on when 50% or more of the veligers were at that stage.

The veligers removed for observations were returned to their respective containers, and dead veligers were removed and recorded to determine mortality.

The sample size taken every two days for measurements of growth during the experiment was n = 30 from each replicate (container culture). Growth was assessed recording increments of shell length (siphonal length). Larvae were measured using a compound microscope with a calibrated ocular micrometer to the nearest 0.10 µm.

Survival rates were calculated using the number of living larvae at the beginning of the experiment and end of the experiment.

ANOVA was used to determine if shell lengths were significantly different for veligers grown in different months. Cochran's test was used to test for homogeneity of variances. Tukey's multiple comparison tests of means was used to compare shell length and mortality data. The statistical program STATISTICA was used for the statistical analyses. Significance was assumed when $p < 0.05$.

RESULTS

Development

Figure 1a shows the evolution of developmental characteristics in the experimental cultures. Veligers of *S. gigas* grown in March, April, May, and September cultures developed two velar lobes and 1.5 whorls at hatching, adult heart appear at 11th days old, beginning six lobes and 3.0 whorls at 13 days, proboscis and radula appear at 19 days, and between 20 to 23 days, respectively, and reached settlement at 29 days.

Veligers from June, July, and August cultures developed four velar lobes and 2.0 whorls at hatching, adult heart appear at 7 days, beginning six lobes and 3.5 whorls at 11th day, proboscis and radula appear at 17 and 21 days, respectively, and reached settlement at 26 ± 1 day (Figure 1b).

Fifty percent or more of the veligers in these cultures were in the following characteristics: 2, two velar lobes and 1.5 - 2.0 whorls for 3 days; 4a, four lobes, 2.0 - 3.5 whorls without heart for 5 to 6 days; 4b, four lobes, 2.0 - 3.5 whorls with heart for 3 to 5 days; 6a, six lobes, 3.0 - 4.0 whorls without proboscis for 4 to 6 days; 6b, six lobes, 3.0 - 4.0 whorls with proboscis for 3 to 4 days; 6c, six lobes, 4.0 - 4.5 whorls without radula for 3 to 5 days; 6d, six lobes, 4.0 - 4.5 whorls with radula for 2 to 5 days; 6e, Settlement.

Larvae from June, July and August cultures did not develop two velar lobes-1.5 whorls, two velar lobes-2.0 whorls and six lobes-3.0 whorls. Veligers did not developed settlement beyond of 27 days. In general, the development from these months was much better that the other group of months (March, April, May, and September).

Growth

Table 1 shows initial shell length (at hatching), final shell length, growth rate, and mortality. At hatching the larvae showed at shell length from 293 µm to 303 µm and average of 298 ± 5.21 µm (n = 630 larvae).

Larvae showed an average growth rate between 31.36 µm/day and 42.80 µm/day and reached an average final shell length at metamorphosis of 941 µm – 1284 µm.

Figure 2 shows the growth for larvae of *S. gigas* cultured from March to September. Larvae from August and September showed the highest growth with 1,284 and 1,276 µm, respectively. Statistical difference was observed between growth of larvae from June and July. No differences were found for larvae growth of March, April and May and for larvae grown in August and September (Table 1).

Mortality

Three mortality periods were observed during the larval cycle: first, between 11 to 13th days, second at 21 days, and the last between 25 to 27 days. In the first period, registered percentages were more than 20%, observed the higher values in May, June, and July with 30%, 31% and 30%, respectively, whilst the lowest were founded in March (26%) and September (25%) cultures.

During the second period, the lowest percentages were registered in June and August with 8% and 9%, respectively. Highest values were observed in cultures from April (13%) and September (12%).

For the third period, culture of March present mortalities of 34%, April culture registered lowest mortality with 12%; May, June, and July with 17%, and August and September cultures 25%. Tukey test demonstrate significant differences between month cultures (p = 0.001).

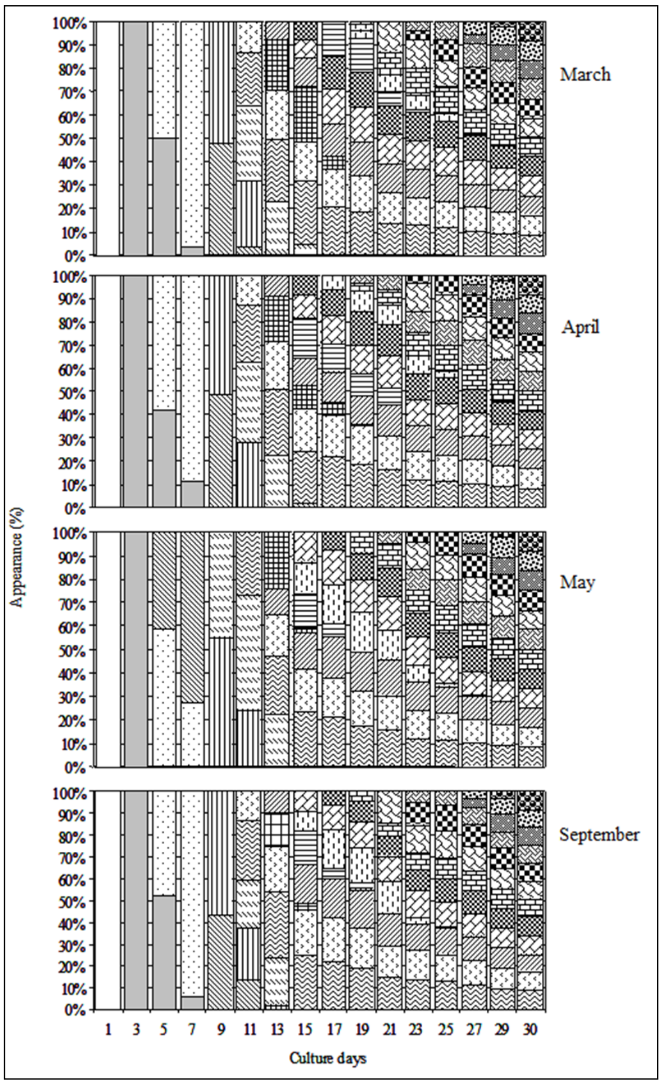

Figure 1a. Developmental characteristics evolution of experimental cultures realized in March, April, May and September, for *S. gigas* from Alacranes Reef.

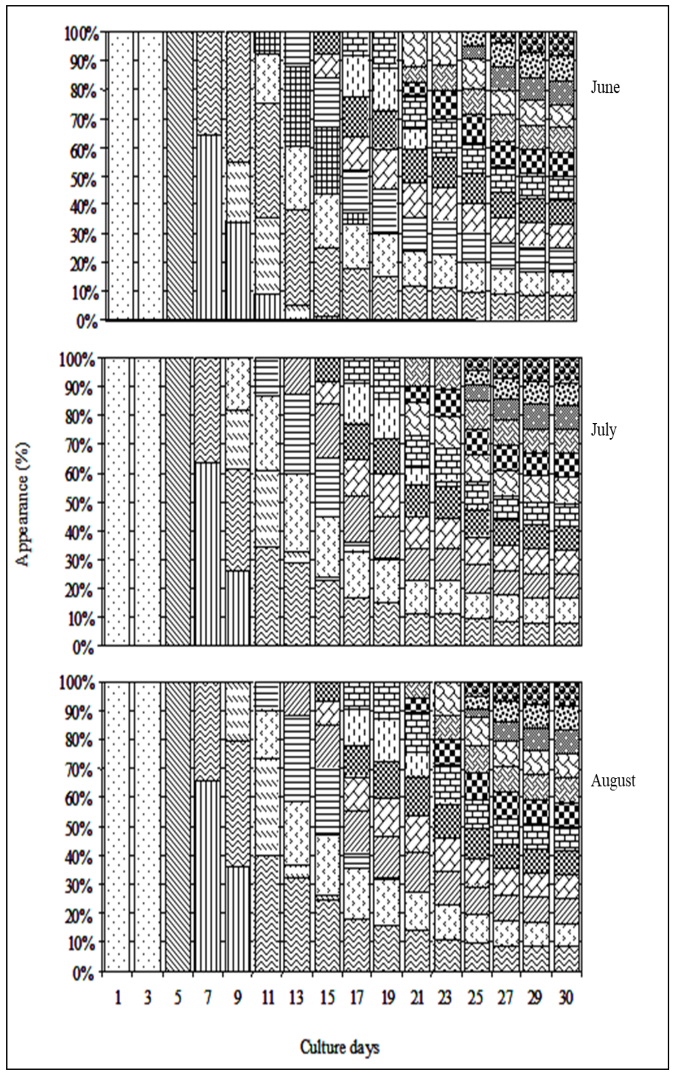

Figure 1b. Developmental characteristics evolution of experimental cultures realized in June, July and August, for *S. gigas* from Alacranes Reef.

Table 1. Growth of shell length for the larvae of *Strombus gigas* in the cultures realized from March to September. Shell length at hatching, final shell length, average growth rate at 30 days and total mortality.

Culture Months	Growth			Total Mortality (%)
	Initial (µm)	Final (µm)	Growth rate (µm day-1)	
March	300 ± 2.63	941 ± 11.92 [a]	31.36 ± 3.60 [a]	71 [a]
April	297 ± 1.89	953 ± 11.81 [a]	31.76 ± 2.54 [a]	53 [b]
May	303 ± 3.01	972 ± 11.93 [a]	32.40 ± 2.72 [a]	57 [b]
June	293 ± 2.30	1189 ± 22.83 [c]	39.63 ± 3.01 [b]	56 [b]
July	295 ± 0.98	1211 ± 21.65 [c]	40.36 ± 2.96 [c]	57 [b]
August	301 ± 2.48	1284 ± 18.54 [d]	42.80 ± 2.96 [c]	61 [b]
September	303 ± 2.32	1276 ± 20.95 [d]	42.53 ± 2.96 [c]	62 [b]

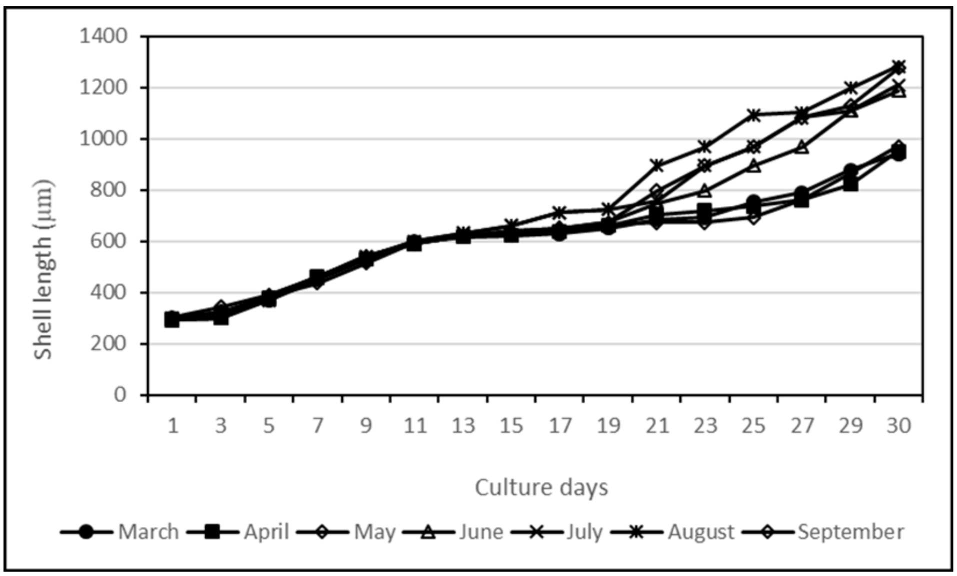

Figure 2. Shell length growth curves for all cultures of *Strombus gigas* larvae from Alacranes Reef.

DISCUSSION

At hatching the larvae of March, April, May, and September have 1.5 shell whorls and two velar lobes, which is the same as *S. gigas* and *S. costatus* (Brownell 1977, Davis et al. 1993), whilst larvae from June to August presented four velar lobes and 2.0 whorls of the shell, characteristics observed by D'Asaro (1965), Brownell (1977), and Davis et al. (1993) at four or five days.

Brownell (1977) reported that the proboscis appears as early as the 25th day in *S. gigas* and *S. costatus*, and begins to function in grazing on algae within 2 days. In this study, proboscis appears at the 17th day and begins to function within 3 days for the larvae of June, July, and August. D'Asaro (1965) and Brownell (1977) observed the metamorphosis between 28 to 33 days, Sidall (1983) in 20 to 25 days, Davis and Hesse (1983) in 28 days, whilst Corral and Ogawa (1985) obtained the metamorphosis between 25 to 60 days and Ray and Davis (1989), observed this characteristic bewtween 21 to 40 days. In this study, metamorphosis took from 27 to 30 days, with 30% of larvae that had not settled in June.

Larval development in this work demonstrates that larvae of March, April, May, and September are smaller than those obtained ones in June, July, and August.

In this study, the average growth rate varied from 31.26 µm/day to 42.80 µm/day at a density of 200 larvae/L. Ballantine and Appeldoorn (1983) reported an optimal growth rate of 50 - 55 µm/day, when the larval density was 100 larvae/L, Aldana Aranda and Torrentera (1987) reached a growth rate of 40 µm/day at 200 larvae/L, Aldana Aranda et al. (1989) working at same larval density with *S. costatus* larvae obtained 16.7 µm/day, Heyman et al. (1989) report 24 µm/day at a larval density of 500 - 600 larvae/L. Davis et al. (1993) working at a larval density of 100 - 200 larvae/L report a growth rate of 39 µm/day, Domínguez Tec (1993) reached 5 - 13 µm/day at a larval density of 60 larvae/L and García Santaella and Aldana Aranda (1994) obtained a growth rate of 24 - 37 µm/day, when the larval density was 275 larvae/L. So, these results only could be compared using larval density of 60 to 600 larvae/L. The results obtained in this work demonstrate that the growth rate was similar or superior to the reported average, the similar or even better, to the reported ones by the investigators of Mexico.

In this work, the *S. gigas* larvae reached a mean shell length at metamorphosis between 941 to 1284 µm. This value is similar to literature data for other *Strombus* spp., except for the values reported by Brownell's which are above the average (2,200 µm at metamorphosis). Ballantine (1981) reported 900 µm for *S. gigas.* García Santella and Aldana Aranda (1994) obtained a maximal shell length of 800 µm for *S. gigas* larvae. Larval growth varies according to the culture conditions and also to the author's criteria. Nevertheless, in this work, the best growths were obtained in August and September.

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