# Atypical Reproductive Cycle of the Queen Conch *Strombus gigas* (Mollusca: gastropoda)

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#### ABSTRACT

The present study of the reproductive cycle of *Strombus gigas* at San Andres, Providence y Santa Catalina, Colombia revealed a problematic situation in this population. Samples were obtained by a cooperation program "Estrategias reproductivas del Caracol pala *Strombus gigas* en el Caribe Insular colombiano" No. 003/abril, 2003, between Departamento Archipiélago de San Andrés, Providencia y Santa Catalina, Colombia and Laboratorio de Biología y Acuacultura de Moluscos, CINVESTAV IPN Mérida, Yucatán, Mexico. 311 organisms were analyzed from February 2003-to January 2004, with a shell length  $\geq 230$  mm. Samples from visceral mass and gonad were taken and processed for histological study. Gametogenesis was observed all year round but at a low percentage. Mature stage was observed only in August-September with percentages  $\leq 10\%$ . Such a detrimental situation may be related to an intense and generalized putative sporozoan infection detected in the sampled *S. gigas* population. The parasite, apparently a *Coccidian* was found in the digestive gland of every sampled organism throughout the year, infecting from 70% to 100% of the digestive gland alveoli, with a frequent total invasion of every alveolar cell. This infection may be responsible for the low intensity of maturity and scarce spawning stages registered at San Andres Archipelago.

However, the actual knowledge of spatial and temporal variations of the reproductive cycle of *S. gigas* and of its parasitic status is not sufficient to propose an efficient management for its sustainable exploitation. Complementary studies appear necessary and urgent to understand its biological status throughout the reproductive cycle. Meanwhile, a mandatory minimal catch size could be a lip thickness > 7 mm in order to protect the endangered reproductive stock of *S. gigas* at San Andrés. The level of protection will depend on an efficient survey and protection of the populations and a high enforcement of regulations to control exploitation and reduce illegal fishing.

KEY WORDS: Queen conch, Strombus gigas, reproduction, anomolies, Colombian

# Anomalias en el Ciclo Reproductor de Strombus gigas (Mollusca: gastropoda)

En este trabajo se presentan las anomalías observadas en el ciclo reproductivo de *Strombus gigas* del Archipiélago de San Andrés, Colombia en un ciclo anual para organismos de talla igual o superior a 230 mm de longitud sifonal. Las muestras biológicas fueron obtenidas a través del convenio "Estrategias reproductivas del Caracol pala *Strombus gigas* en el Caribe Insular colombiano" de cooperación técnica No. 003/abril, 2003, celebrado entre el Departamento Archipiélago de San Andrés, Providencia y Santa Catalina, Colombia y el Laboratorio de Biología y Cultivo de Moluscos CINVESTAV-IPN, Mérida, México. Se analizaron 311 organismos colectados de febrero de 2003 a enero de 2004.V Las muestras de masa visceral y gónada fueron fijadas por histología. Los organismos analizados presentaron un rango de tallas entre 200-290 mm de longitud sifonal y grosor del labio de 1-30 mm. Los resultados muestran gametogénesis todo el año pero con frecuencias bajas no mayores al 50% en agosto y noviembre. El estadio de madurez se presenta solo en agosto-septiembre y en porcentajes muy bajos, menores a 10%. Estas anomalías que presenta el proceso de madurez de la gónada pudieran tener relación con el alto porcentaje de incidencia de parasitismo que fue observada en la glándula digestiva. El parásito, aparentemente un *Coccidio* localizado en la glándula digestiva de todos los organismos muestreados en un ciclo anual, infectando del 70% al 100% de los alvéolos glandulares de la glándula digestiva.

La falta de estudios sobre el ciclo reproductivo del caracol *Strombus gigas*, sus variaciones espaciales y sus anomalías dificultan el planteamiento de medidas de regulación *ad hoc y per se* al ciclo de vida de esta especie, para su uso sustentable. Con base en los resultados de este estudio se propone una revisión cuidadosa de esta especie para conocer las causas que están afectando el proceso de madurez de esta especie y reajustar la veda para *S*.*gigas* en Colombia a fin de proteger el delicado stock de organismos que alcanzan la madurez.

En el intervalo de este segundo estudio es necesario no permitir la captura de organismos con grosor de labio inferior a 6 mm.

PALABRAS CLAVES: Caracol pala, *Strombus gigas*, ciclo reproductivo, anomalías, Colombiano

## **INTRODUCTION**

The queen conch, *Strombus gigas* Linne, 1758 is a large gastropod of commercial importance in the Caribbean. With a distribution range from Venezuela to Florida and the Bahamas, including the lesser and Greater Antilles, it is known with several common names of local usage.

Due to the high fishing pressure that is exerted upon most of its populations, several stocks have been reduced to levels where the population can no longer recover (Appeldoorn 1994a), and commercial fishing is no longer feasible.

Due to high demand, vulnerability and overexploitation of local stocks, the species has been included in the Convention on International Trade of Endangered Species of Wild Fauna and Flora since 1992 (CITES, 2003), and in 1994 it was added to the International Union for the Conservation of the Nature's Red List (IUCN). Ardila, *et al.* (2002) included *S. gigas* in the red book of marine invertebrates of Colombia as a vulnerable species because of over-fishing and loss of its habitat.

Along the Caribbean region, different management strategies are applied to limit exploitation. Strategies that focus on the resource are: minimum legal size expressed in length of shell, meat weight or lip thickness, catch quotas, temporal fishing bans, reproductive bans. Strategies to limit fishing effort have been: limited number of fishers, number of conchs per fisher, boats per day; restriction of capture by depth, restrictions to the use of scuba or hookah diving equipment. Strategies to protect reproductive stocks or populations in recovery are the establishment of closed areas to fishing activity and the establishment of Marine Protected Areas.

Management strategies are based on a wide range of information sources, from commercial catches statistics to biological sampling. From CITES recommendations, some sort of stock assessment should be made, together with population dynamics which include variations of population structure in time and space, reproductive periods and growth parameters.

*S. gigas* in Colombia is the second marine invertebrate commercially more harvested after the lobster. Castro-Gonzalez (2003) indicated that the harvest quote of Conch for 2001 was of 96 tons.

The fishing pressure at San Andres has caused the diminution of the populations of *S. gigas*, forcing to the establishment of regulatory measures as:

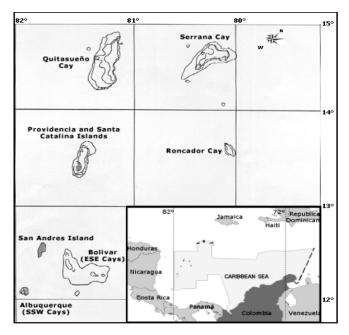
- i) Authorized catch techniques (free snorkel);
- ii) Minimum size of capture evaluated by weight at 100g of clean meat (only muscle) or 225g

uncleaned meat (with mantle, viscera and operculum);

- iii) Fishing prohibition during 01 June 31 October. and
- iv) Total closure of the fishery to Quitasueño Cay (Resolution No. 000179 of 05 May, 1995). The objective of this work is to report anomalies observed in the reproductive cycle of *S. gigas* at San Andres and propose management measures for the queen conch fishery of Colombia.

#### Sampling Site

Strombus gigas was sampled from southwestern of San Andres, Colombia (Figure 1) between the  $12^{\circ}32'N 81^{\circ}$ 42'W (San Andrés island),  $12^{\circ}24'N 81^{\circ}28'W$  (Bolivar Cays) and  $12^{\circ}10'N 81^{\circ}51'W$  (Albuquerque cays). San Andrés is a barrier reef of 32 km long and covers an area of 255 km<sup>2</sup>, one of the largest coral reefs in America. Bolivar Cays are located at east, southeast of San Andres. They constitute an area of 10 km in length and enclose four cays. Albuquerque cays are located to the southwest of San Andrés. This Archipelago was declared as the Seaflower Biosphere Reserve in November 2000 by UNESCO's Program. (IGAC, 1986).



**Figure 1.** Sampling site of *Strombus gigas* at Archipelago of San Andrés, Providencia and Santa Catalina (SAI), Colombia (Seaflower Biosphere Reserve) (Avila et al., 2005)

Fishermen of Fishingman Place Cove Sea Side cooperative collected conchs from February 2003 to January 2004 of the categories "Roundshell" and "Bradleaf" (shell total length greater to 22 cm). Organisms were sampled monthly and processed for histological

methods. Lip thickness, total length of shell and sex were determined. A sample of  $1 \text{ cm}^3$  of the visceral mass and gonad of each specimen was dissected and prefixed 15 days in 10% saline formalin, then samples were preserved in 70% alcohol with glycerin, until their transfer to CINVESTAV IPN in Yucatan, Mexico.

## Histological Methods

The samples were post fixed 7 days in alcoholic Bouin's fixative; rinsed in 70% alcohol; dehydrated in an ascending ethanol series (70%, 96% and 100%); clarified in Clearene<sup>®</sup>, a clearing-solvent of the paraffin formulated with selected blend of terpenes; then, embedded in Paraplast<sup>®</sup> tissue embedding medium (m.p. 56°C), a compound of purified paraffin and plastic polymer of regulated molecular weights. Six microns sections were cut with a MICROM HM340E rotary microtome and mounted on glass slides. Sections were stained using Harris's Hematoxylin and Eosin (HHE<sub>2</sub>), regressive stain method (Howard and Smith 1983). Tissue slides were examined under 100x, 400x magnifications using a Carl Zeiss MC73A light microscope.

## **Stages of Reproductive Cycle**

The gonad development stages were defined following the microscopic characteristics described by Aldana-Aranda *et al.* (2003a, b), who were considering five stages. These stages were reduced to four gonadic stages in this study:

- i) *Undifferentiated*, the gametes may not be identified from the microscopic section,
- ii) *Gametogenesis*, active cell division, mature gametes may or may not be present,
- iii) *Mature*, dominance of mature gametes, although some gametogenesis may be present, and
- iv) *Spawn*, follicles are partially or totally emptied and broken, eggs and sperms are being reabsorbed with the presence of phagocytes.

#### RESULTS

#### **Reproductive stages**

*Undifferentiated* — in both sexes, there are no signs of follicles producing germinal cells. The gonad area was occupied almost in its totality by connective tissue. Numerous dispersed amoebocytes were observed. Few broken follicles can be seen (Figure 2a).

*Gametogenesis* — follicles producing germinal cells were observed dispersed and occupying at least 25% of the gonad area. In females, ovogonias and oocytes groups were observed (Figure 2b). In males, spermatogonia and spermatocytes are present (Figure 2c).

*Mature* — in females, follicles reached an average of  $266.50 \pm 77.15$  mm, their walls are very thin and have anastomosis with each others. Early germinal cells (ovogonias and previtellogenic oocytes) are absent. Eggs fill the follicles, they measured  $168.50 \pm 20.37$  mm with abundant vitelline granules (Figure 2d). Male gonads show anastomosed follicles and occupy the whole gonadic tissue; their diameter is  $114.00 \pm 24.51$  mm. Sperms are the dominant stage, forming a large mass in the center of the follicles of eupyrene, oligopyrene and apyrene sperms. The vases deferent are clearly formed and filled with sperm, although some sections may be empty (Figure 2e).

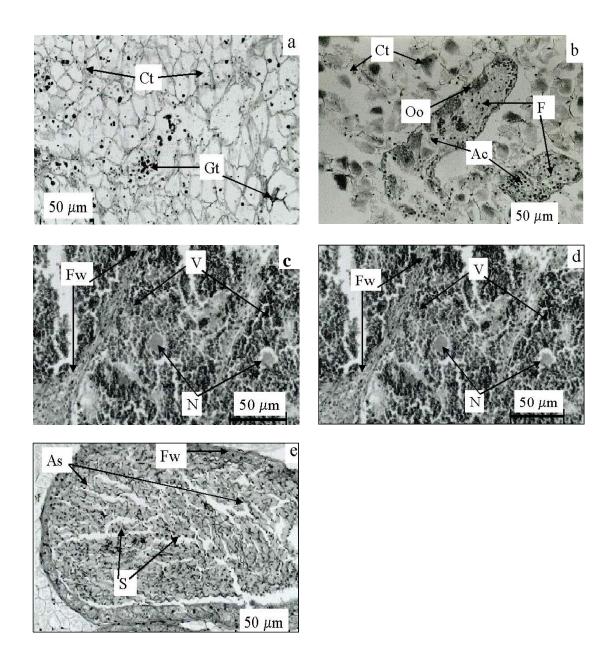
*Spawn* — in females the follicles are almost emptied. Connective tissue starts to form from the outer wall of the gonad and between the gonad and digestive gland. Sparse ovocytes and residual oocytes in re-absorption can be detected with dispersed vitelline platelets, (Figure 2f). In males the follicles continue to anastomose. Sperms are moved to the testis duct, leaving empty spaces in the follicles (Figure 2g).

## **Reproductive Cycle in Males**

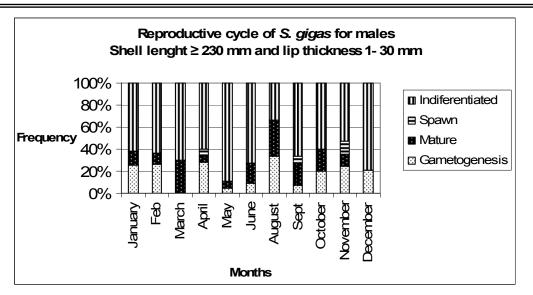
Gametogenesis stage was present throughout the sampling period, with an average of 20%, except in August (33%). Mature stage was also present throughout the year with small percentages, except in March and August (33%). Spawn stage is limited to April, September (7%) and November (10%). Undifferentiated stage was profuse and presented through the year with percentages  $\geq$  60% (Figure 3).

### **Reproductive Cycle in Females**

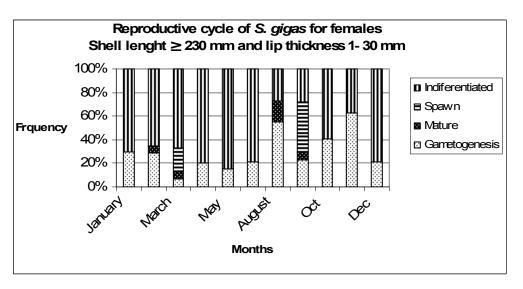
Gametogenesis was present throughout the sampling period, it represented an average of 30%, with slightly higher frequency during August (58%) and November (60%). Mature stage is absent through the sampling period, excepted for August 18%, February, March and September with percentages  $\leq 5\%$ . Spawn stage was no detected, giving place to a long post-spawn, undifferentiated stage throughout the sampling period. The undifferentiated stage was present throughout the year with high percentages  $\geq 60\%$ , except in August and September. During which a minimum of 27 % was observed (Figure 4).



**Figure 2.** Photomicrographies of *Strombus gigas* gonad. **a**) Rest stage. **b**) Gametogenesis in females. **c**) Gametogenesis in males **d**) Mature females **e**) Mature males **f**) Spawn in females. **g**) Spawn in males. Ac: Accessory cells; As: Atypical spermatocytes; Ct: connective tissue; F: Follicle; Fw: Follicle wall; Gt: Germinal tissue; N: Nucleus; O: Ova; Oo: oocytes; V: vitellum; S: spermatozoids; Sg: Spermatogonias; S1: Spermatocytes; Sd: Spermatides (Aldana Aranda, *et al.* 2003).



**Figure 3.** Reproductive cycle of *Strombus gigas* for males with a length shell ≥230mm from San Andres, Colombia



**Figure 4.** Reproductive cycle of *Strombus gigas* for females with a length shell ≥230mm from San Andres, Colombia

## DISCUSSION

Gonad development behaviour of *S. gigas* in this locality showed harms to reach maturity. The reproductive season of *S. gigas* in the Caribbean has been obtained from observations of reproductive behavior: pairing, copulating and egg-laying that not include gonad histological results. Stoner *et al.* (1992) reported from Bermuda a reproductive season from May to September. In the Florida Keys, the reproductive season was between mid-January to mid-September. In Venezuela, it was from mid-March to mid November. Pérez Pérez and Aldana Aranda (2000) reported a reproductive period of *S. gigas* from May to September in Alacranes reef. Stoner *et al.*, (1992) said there was no apparent trend related to latitude in beginning,

end or length of reproductive season for the queen conch from Bermuda to Venezuela. Geographic comparisons of seasonality in reproduction must be interpreted cautiously due to different methods, frequency and number of observations and different habitat types. Baqueiro-Cárdenas and Aldana-Aranda (2000) indicated that species with an ample geographic distribution exhibit a variety of reproductive patterns for every gonad development stage, depending on the percentage and duration.

Aldana-Aranda *et al.* (2003a, b) mentioned that *S. gigas* exhibits different reproductive patterns, in various localities when analyzed by histological studies. For *S. gigas* from Chinchorro bank they reported gametogenesis and undifferentiated stages long and low. Mature stage

was observed in females and males with an average percentage of 35% from May to September.

*S. gigas* at Chinchorro Bank from Mexico presented gametogenesis during spring and summer, and apparently continued during early autumn, with the presence of mature organisms, that extend from spring to summer. Lastingness and intensity of the gonad stages are a function of temperature and food availability according to several authors (Webber 1977, Bayne 1978, Sastry 1979, Fretter 1984, Mackie 1984), as well as of other environmental factors (Bricelj and Malouf 1980, Syed-Shafe 1980, Jaramillo *et al.* 1993).

For *S. gigas* at Chinchorro one or several of these factors may be necessary to induce a massive spawn, as only a fraction of the mature population spawned, with an increasing number of mature organisms towards fall. Spawn may be induced by several factors, such as temperature (Hines 1979, Stoner *et al.* 1992). Spawning has been reported from March to October at different localities in the Caribbean, being more intense from May to August (Berg 1981, Hesse 1976, Orr and Berg 1987, Pérez Pérez and Aldana Aranda 2000).

A higher frequency of gametogenic and mature organisms after the spawning peak of July is evidence of a rapid gonad recovery during summer, which is not the case during fall and spring. Slow gonad recovery is evidenced by increasing frequency of organisms at an undifferentiated stage during September.

Aldana Aranda, *et al.* (2003) studied the gonadic development of *S. gigas* at Alacranes reef from Mexico. They observed for *S. gigas*, on Alacranes reef, mature and spawn stages short and low, limited to June, August, and September ( $\leq 10\%$ ). It could be interpreted as no accumulation of ripe gametes. Post-spawn was long and high, and gametogenesis abundant. Aldana Aranda, *et al.* (2003) mentioned that the gametogenic period for *S. gigas* presented differences between Alacranes reef and Chin-chorro bank, even though the gonad samples were taken during the same period. Therefore, it can be ascertained that the reproductively activity was controlled by temperature and photoperiod conditions.

In this study, S. gigas at San Andres underwent gametogenesis throughout the year but at a low percentage. Mature stage was observed only in August-September with percentages  $\leq 10\%$ . These first results would suggest a fast gametogenic activity without accumulation of ripe Organisms from this locality exhibited an gametes. undifferentiated stage profuse throughout the year. For a preliminary study with a few samples of S. gigas per month, Avila et al (2005) did not observed mature stage from February to August for shell length organisms of 200 mm and few gametogenesis  $\leq 10\%$  except in March (30%). Baqueiro et al. (2005) for S. gigas of Alacranes reef, Mexico and San Andrés, Colombia, found in the digestive gland an intense and generalized sporozoan infection detected during reproductive studies of S. gigas populations from these localities. The parasite apparently was a coccidian found in the digestive gland of every sampled organism throughout the year, infecting from 70% to 100% of this tissues. This infection could be responsible for the low intensity of gonadal maturity registered at San Andres, Colombia, and Alacranes reefs, Mexico. Kennedy (1983) mentioned that stress conditions (e.g. poor food quality, shell or gill damage) may influence sexual condition in oyster population, and suggested that an increased proportion of undifferentiated stage or males in an area may indicate the presence of an environmental stressor.

From the results presented here, complimentary studies of the gonadal development, and the presence of parasites in the digestive gland of *S. gigas*, are recommended throughout the Caribbean Sea. The aim of a systemic study is to identify which environmental factors influence the gonadal development and spawning periods of the queen conch, as well as the possible role of parasitism on the actual low reproductive success of the species in several populations.

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