

**Preliminary Data of the Gametogenic Cycle of *Strombus gigas*
from Archipelago of San Andrés, Providencia and Santa Catalina,
Colombia (Seaflower Biosphere Reserve)**

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ABSTRACT

Preliminary results of the gonad development stages of *S. gigas* are presented from February to August 2003, at the Archipelago of San Andrés, Providencia and Santa Catalina, Colombia (research project Colombia-Mexico "Estrategias reproductivas del Caracol pala *Strombus gigas* en el Caribe insular colombiano"). This Archipelago was declared the Seaflower Biosphere Reserve in November 2000 by UNESCO's Programme on Man and the Biosphere (MAB). Ten organisms were sampled monthly and processed by histological methods. Gonad development stages were described. Rest stage was observed from February to May (20 %, 20 %, 40 % and 10 %, respectively). Organisms in gametogenesis stage were observed from March to August (50 %, 10 %, 10 %, 10% and 30 %, respectively). Mature stage organisms were not observed during this study. Spawn stage was detected only in April (10 %). Post spawn stage was observed from February to August with 30 % to 90 %. These first results would suggest a fast gametogenic activity without accumulation of ripe gametes.

KEY WORDS: *Strombus gigas*, queen conch, gametogenesis, Colombia

**Datos Preliminares del Ciclo Gametogénico de *Strombus gigas* del
Archipiélago of San Andrés, Providencia and Santa Catalina, Colombia
(Seaflower: Reserva de la Biosfera)**

Son presentados resultados preliminares de los estados de desarrollo de la gónada de *S. gigas* estudiados de febrero a Agosto 2003, en el Archipiélago de San Andrés, Providencia and Santa Catalina, Colombia (proyecto de investiga-

ción conjunto Colombia-México "Estrategias reproductivas del caracol Pala *Strombus gigas* en el Caribe insular colombiano"). Este Archipiélago fue declarado Reserva de la Biosfera Seaflower en noviembre de 2000 por el Programa el hombre y la biosfera (MAB) de la UNESCO. Diez organismos fueron muestreados mensualmente y procesados por métodos histológicos. Se describieron los estados de desarrollo gonádico. El estado de reposo fue observado de febrero a mayo con porcentajes de 20, 20, 40, y 10, respectivamente. Organismos en estado de gametogenesis fueron observados de marzo hasta agosto (50 %, 10 %, 10 %, 10 % y 30%, respectivamente). No se observaron organismos en estadio de madurez durante éste estudio. El estado de evacuación fue detectado solamente en abril con 10 %. El estado de post evacuación fue observado desde febrero hasta agosto con porcentajes de 30 hasta 90. Estos primeros resultados sugerirían una actividad gametogenica rápida sin acumulación de gametes maduros.

PALABRAS CLAVES: *Strombus gigas*, caracol, gametogenesis, Colombia

INTRODUCTION

The queen conch *Strombus gigas* Linnaeus, 1758 (Gastropoda: Caenogastropoda: Strombidae) is a marine resource of commercial importance in several Caribbean countries supporting a high fishing pressure that has reduced its populations (Aldana-Aranda 2003). Since 1992, *S. gigas* has been included in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES), 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 in the Red Book of marine invertebrates of Colombia a *S. gigas* as a specie vulnerable (VU A2d) due to overfishing and loss of its habitat. Colombia, as a member of CITES, controls the international market of *S. gigas* and contributes to sustainable management of *S. gigas* in this region (Reyes and Santodomingo 2002).

In Colombia, *S. gigas* is the second most commercially harvested marine invertebrate after lobster, due to its size, attractive shell, flavourful meat, and high demand in the North American market. Ospina-Arango et al. (1996) indicated that the main conch harvest is at San Andrés, Providencia and Santa Catalina (SAI). Castro-Gonzalez (2003) indicated that the harvest of conch in 2001 was 96 tons, however, 100 tons of conch were harvested this year. The fishing pressure at SAI has caused the diminution of the populations of *S. gigas*, forcing to the establishment of regulatory measures as:

- i) Catch techniques authorized (free diving to lung);
- ii) Minimum size of capture in 100 grams meat clean (without mantle, visceral, and operculum) or 225 grams no meat clean (with mantle, visceral and operculum);
- iii) Fishing prohibition during the reproduction season (01 June - 31 October); and
- iv) Total closures of the fishery to Quitasueño Cay (Resolution No. 000179 of 05 May, 1995).

S. gigas from Colombia had been studied by Chiquillo-Espitia et al.

(1997), Ospina-Arango et al. (1996), Márquez-Pretel et al. (1994), Márquez-Pretel (1993), Arango y Márquez-Pretel (1993), García et al. (1992), y García (1991). These authors studied capture data and some biological aspects. Chiquillo-Espitia et al. (1997) characterized the gonad development stage of *S. gigas* of SAI by macroscopic scale. The reproductive season of *S. gigas* at Santa Marta begins in April and ends in July (Botero 1984), for San Bernardo Archipelago beginning in June to January (Lagos-Bayona et al. 1996) and for SAI, from April to September (Márquez-Pretel et al. 1994) and from mid-June to October (García et al. 1992). The inconsistency of data on the reproductive cycle of *S. gigas* is due to different methods used to observe reproductive behavior: pairing, copulating, egg-laying and/or presence of egg masses.

The gonadal development of *S. gigas* of SAI in this study was analyzed by a histological method describing the gonad development stages from February to August 2003.

MATERIALS AND METHODS

Sampling Area

Strombus gigas was sampled from southwestern of SAI, Colombia (Figure 1) between the 12°32'N 81°42'W (San Andrés island), 12°24'N 81°28'W (Bolivar Cays) and 12°10'N 81°51'W (Albuquerque cays). *San Andrés* is a barrier reef of 32 km long and covers an area of 255 km², making it one of the largest coral reefs in the Americas. It is identified as a major site of coral and fish diversity and is considered a biodiversity "hotspot" with 27 km² and an official population of 70.657 to 2002 (The Ocean Conservancy, 2003). *Bolivar Cays*, is located to 29 km to the East Southeast of San Andrés Island, constituted an area of 10 km of length, enclosed four small cays totally coralline and sandy, with 24 km of perimeter and 9 m of depth. In the east of these cays, there is a formation of white sandy that finish in a reef of 5 km in length (IGAC 1986). *Albuquerque cays*, is located to 40 km to the southwest of San Andrés Island, within a cleared coralline set of 23 km of perimeter. Its length is of 8 km. In the West of this cays are sandy, rocks and shallow area of 1 on to 2 meters, alternating with depths of 7 meters and an atoll lagoon (IGAC 1986).

Organism Sampling

Fishermen of the cooperative "Fishingman Place Cove Sea Side" collected conchs from February to August 2003, except in July by Claudette hurricane. Ten organisms between adult and subadult were collected monthly, in the categories Roundshell and Broadleaf (shell total length greater to 22 cm). *Roundshell*: youthful or subadult, that does not even have the formed lip (commonly captured); and *Broadleaf*: with formed lip (always captured).

Lip thickness, total length of shell, and sex were determined. Sample of 1 cm³ of the visceral mass was dissected (gonad and digestive gland) and prefixed 15 days in 10 % saline formalin (sea water with sodium borate), then samples was preserved in 70 % alcohol with glycerin, until their transfer to mexican laboratory.

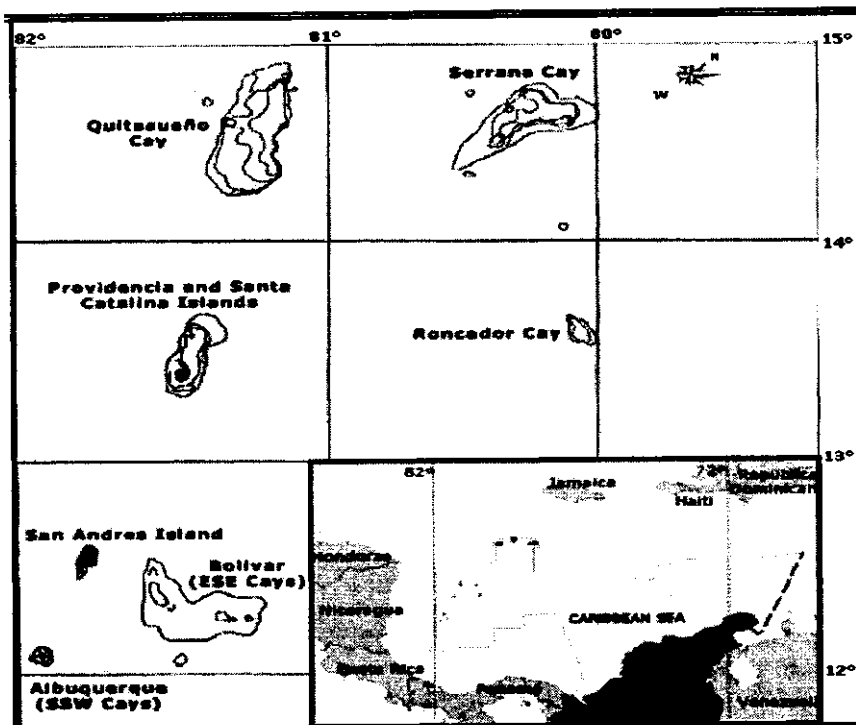


Figure 1. Sampling site of *Strombus gigas* at Archipelago of San Andrés, Providencia and Santa Catalina (SAI), Colombia (Seaflower Biosphere Reserve)

Histological Procedure

The samples were postfixed seven days in alcoholic Bouin's fixative; rinsed in 70 % alcohol; dehydrated in an ascending ethanol series (70 %, 96% and 100 %); clarified in Clearene[®], a clearant-solvent of the paraffin formulated with selected blend of terpenes. Later, embedded in Paraplast[®] 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 (HHE₂), regressive stain method (Howard and Smith 1983). Tissue slides were examined under 100x, 400x magnifications using a Carl Zeiss MC73A light microscope.

Establishment of Stages of Development

The gonad development stages were defined following the microscopic characteristics described by Aldana-Aranda et al. (2003a, b), who considering five stages: *Rest* = the sex is not yet apparent cytologically; *Gametogenesis* = begins gametogenesis, gametes in first stage may or may not be present; *Mature* = presence of mature gametes, number of gametes increases and there is follicle more numerous; *Spawn* = partially emptied follicles and their wall

start to collapse; *Post Spawn* = wide gonad with a large number of expanded but nearly empty follicles. A few gametes may remain.

RESULTS

Gonad Development Stages

Rest — In both sexes, there are no signs of follicles producing germinal cells. The gonad area was occupied almost in its totality by loose connective tissue. Numerous dispersed amoebocytes were observed. Few broken follicles can be seen.

Gametogenesis — Follicles producing germinal cells was observed dispersed and occupying 25 % of the gonadal area. The follicles with germinal cells in stage of advanced development were detected neighboring to the digestive gland. In females, ovogonia groups were observed near the wall of each follicle, whilst the oocytes were observed toward the lumen. In male, the spermatogonia, first order spermatocytes and second order spermatocytes is located near the wall of the follicles, whilst the apyrene and oligopyrene spermatozoa occupied the lumen.

Spawn — Gonad is dominated by swollen, bulging, and anastomose follicles that occupied most gonad area. Loose connective tissue occupies between 10 % and 50 % of gonad area. The follicles neighboring to the digestive gland were detected in direct contact with this gland. In females, the follicles contained principally eggs with its nucleus clear and with an abundance of vitelline granules. In males, we observed vases deferent with a wall constituted of columnar cells clearly formed and filled or semi-filled with spermatozoa.

Post spawn — In both sexes, 90 % of the gonad area is occupied by loose connective tissue. There are follicles broken and some germinal cells in different stages of development. Numerous phagocytes are presented around of the follicles.

Reproductive Cycle

Females — The rest stage was presented throughout March (40 %) to May (20 %). Gametogenesis stage is limited to March (60 %) and minimum of 20 % August. The mature and spawn stages were not detected, giving place to a long Post Spawn stage throughout of the sampling period. Post spawn stage was present from February to June (100%) and May to August (80%), and a minimum of 60 % during April (Figure 2).

Males — The rest stage was present during February and April with 40 %. Gametogenesis stage was present throughout the sampling period, with 40 % in March and August, and 20% from April to June. Mature stage was not present. Spawn stage is limited to April (20%). Post Spawn stage presented a peak of 80 % on May and June, and a second peak of 60 % in February, March and August. Minimum was observed during April (20 %) (Figure 3).

Both sexes — Rest stage was observed from February to May with a peak in April (40 %) and a lower percentage in May (10 %). Gametogenesis stage

was presented throughout the sampling period with two peaks: March (50 %) and August (30 %). The mature stage was not observed. Spawn stage was limited to April with 10 %. Post Spawn stage presented three high peaks: June (90 %), February and May (80 %) and August (70 %); with a minimum in March (30 %) (Figure 4).

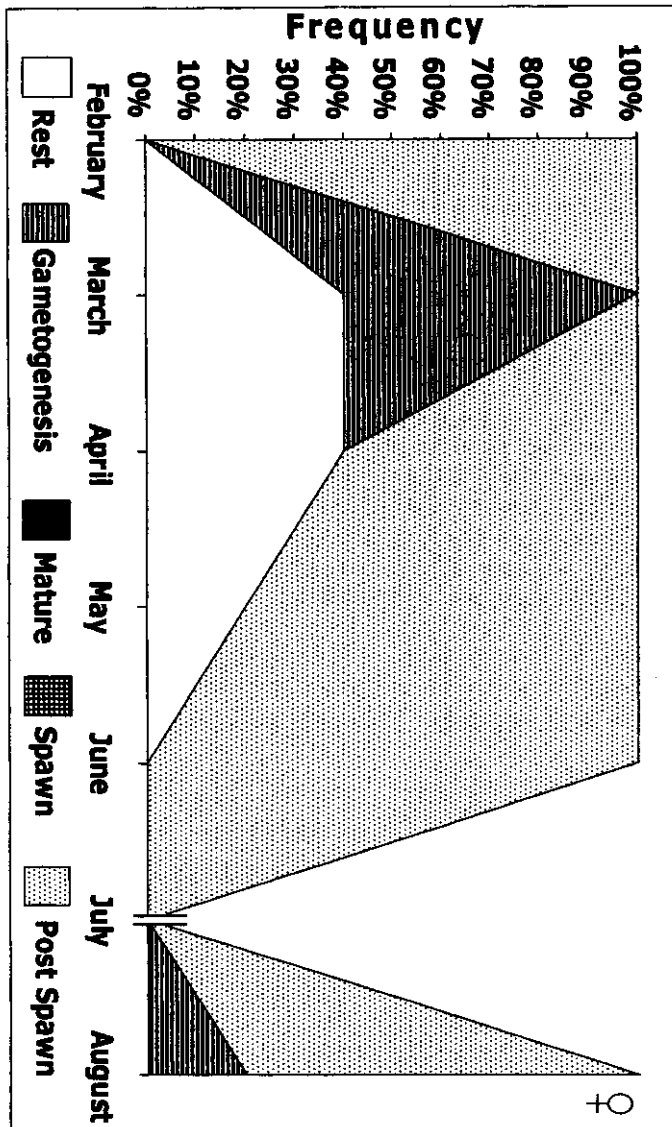


Figure 2. Percentage of females *Strombus gigas* in the different reproductive stages from San Andrés, Colombia

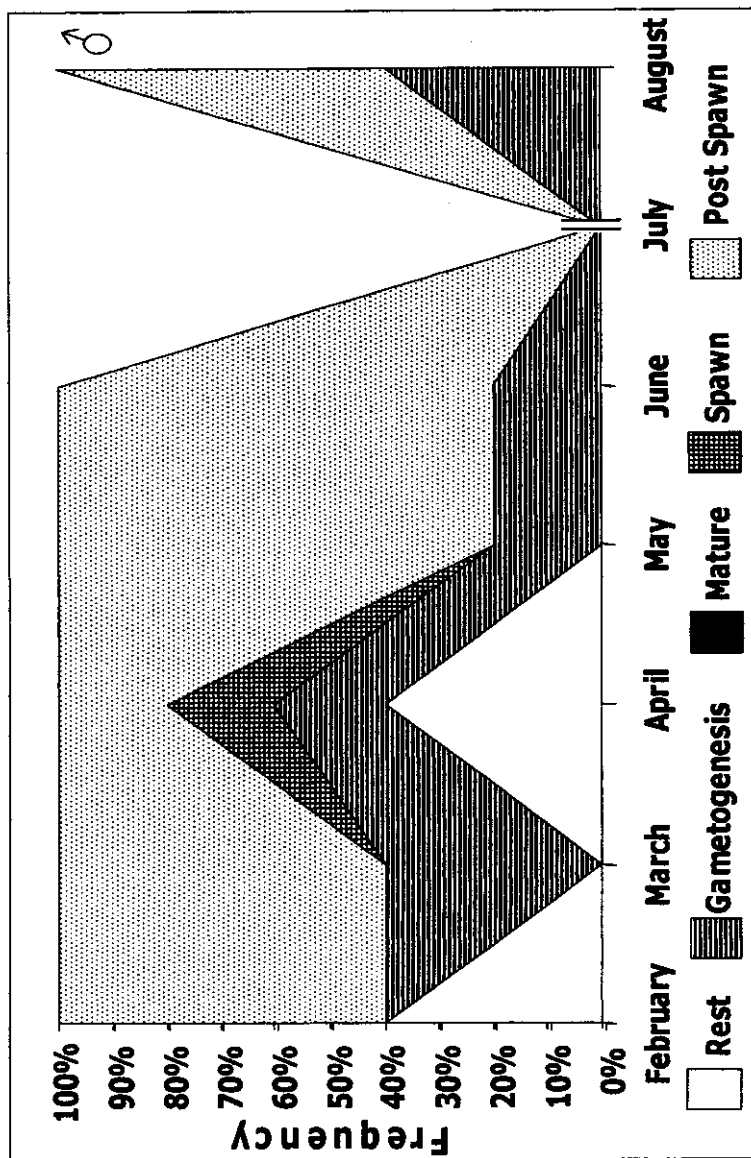


Figure 3. Percentage of males of *Strombus gigas* in the different reproductive stages from San Andrés, Colombia

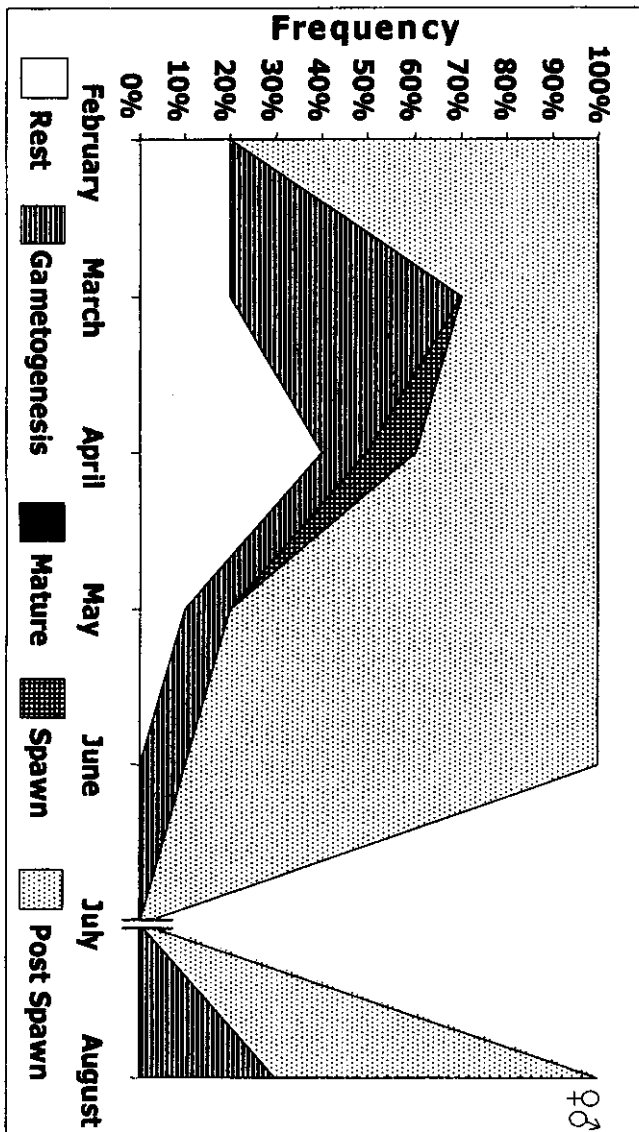


Figure 4. Percentage of both sexes of *Strombus gigas* in the different reproductive stages from San Andrés, Colombia

DISCUSSION

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 (Table 1). The reproductive season of *S. gigas* at SAI Colombia begins in April and ends in September (Márquez-Pretel et al. 1994); or from mid-June to October (García et al. 1992). Based in these studies of *S. gigas*, Colombia established in 1995 a temporal ban from June 01 to October 31 (Resolution No. 000179).

Baqueiro-Cárdenas and Aldana-Aranda (2000) indicated that species with an ample geographic distribution exhibit a variety of reproductive patterns for every gonadal development stage, depending on the percentage and duration. Aldana-Aranda et al. (2003b) indicated that *S. gigas* have different reproductive patterns. In this study, gametogenesis and resting stages were long and low. Mature stage was absence; could be interpreted as no accumulation of ripe gametes. Spawn stage was short and low. Post spawn was long and high (Table 2).

Other studies that also were done by histology of the gonad (Aldana-Aranda et al. 2003a, b) observed a similar reproductive pattern between gonad development stages of *S. gigas*, but with an incompatibility on the beginning, as to spawn and post spawn stages of *S. gigas* in the three locations. Apparently, *S. gigas* from San Andres Island, Chinchorro Bank and Alacranes reef present the same date of beginning for rest, gametogenesis and mature stages (Figure 5). The greater reproductive activity of this species appears when the temperature average of the water begins to rise (> to 24 °C), nevertheless, Stoner et al. (1992) affirms that the reproductive season is less evident in the warmer regions, indicating the probability of reproductive activity throughout the year, whereas the reproductive season is more evident in colder zones or greater latitudes. These first results would suggest a fast gametogenic activity without accumulation of ripe gametes through of whole sampling period, but it is necessary to know the annual cycle for gametogenesis.

Table 2. Gonad development stage of *Strombus gigas* of SAI, following the descriptions of Baqueiro-Cardenas and Aldana-Aranda (2000)

Gonad development stage	duration	percentage	interpretation
Rest	long	low	synchronic and fast rest activity
Gametogenesis	long	low	fast gametogenic activity
Mature	-	absence	no accumulation of ripe gametes
Spawn	short	low	asynchronic spawn
Post spawn	long	high	synchronic and slow post spawn activity

Table 1. Reproductive season of *Strombus gigas* based in observations of reproductive behavior: pairing, copulating, egg-laying and/or presence of eggs mass.

J	F	M	A	M	J	J	A	S	O	N	D	Duration months	Geographical coordinates	Location	Reference
												4.5	32°20' N, 64°45' W	Bermudas	Berg et al., 1982
												4.5	24°40' N, 82°00' W	Florida	Davis et al., 1984
												7	23°35' N, 78°05' W	Bahamas	Sloner et al., 1992
												5.5	23°35' N, 78°05' W	Bahamas	Wicklund et al., 1991
												9	22°30' N, 89°40' W	Mexico	Perez-Perez and Aldana-Aranda, 2002
												7	21°30' N, 72°15' W	Turks y Caicos	Davis et al., 1987
												8	18°15' N, 65°00' W	Islas Virgenes	Randall, 1964
												8	18°15' N, 68°30' W	Puerto Rico	Appeldoorn, 1988
												5.5	17°05' N, 82°30' W	St Kitts/Nevis	Wilkins et al., 1987
												6	13°00' N, 81°00' W	Colombia*	Marquez-Pitel et al., 1994
												4.5	13°00' N, 81°00' W	Colombia*	Garcia et al., 1992
												4	11°30' N, 74°05' W	Colombia†	Balero, 1984
												8	9°45' N, 79°50' W	Colombia‡	Lagos-Bayona et al., 1996
												6	11°40' N, 68°30' W	Venezuela	Brownell, 1977.
												7	11°40' N, 68°30' W	Venezuela	Well and Laughlin, 1984.

* Without later date. † Archipelago of San Andrés, Providencia and Santa Catalina. ‡ Santa Marta and † San Bernardo Archipelago.

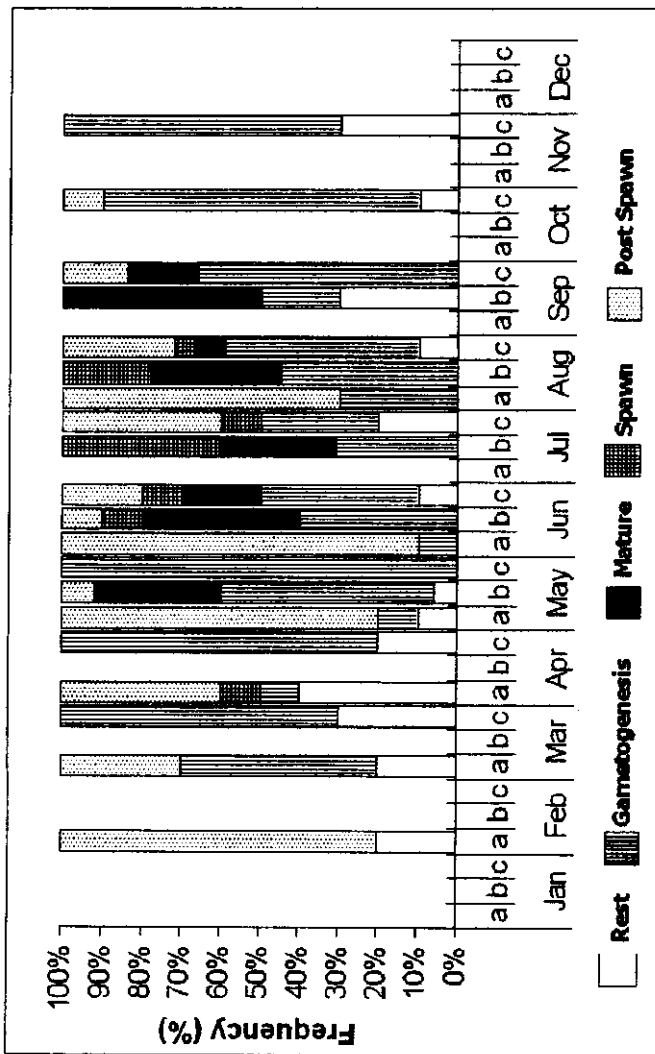


Figure 5. Reproductive patterns of gonad development stages of *S. gigas* by histological procedures for this study and other studies (Aldana-Aranda et al., 2003a, b): a) San Andrés, Colombia; b), Chinchorro Bank, Mexico; c) Alacranes Reef, Mexico

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