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Reproductive aspects of *Acanthurus chirurgus* (Perciformes: Acanthuridae) captured by the artisanal fleet operating in Itamaracá Island, Pernambuco, Brazil

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Summary: This study examined the reproductive biology of *Acanthurus chirurgus* through monthly collections from May 2016 to May 2018. A total of 505 specimens (317 males, 188 females) were analysed, with males having a larger total length than females (32.0 cm vs. 30.8 cm). The highest frequency of individuals was in the 23–25 cm size range. Reproduction occurred from January to July, with peak spawning in February, March and June. The first (L_{50}) and maximum (L_{99}) maturation sizes were 18.0 and 22.1 cm, respectively. Spawning was synchronous, occurring in batches and following an indeterminate fecundity pattern. Spawning frequency was estimated at every 4.2 days (hydrated oocytes) and 3.5 days (post-ovulatory follicles). Batch fecundity ranged from 15403.9 to 48030.9 oocytes, and relative batch fecundity from 2710628.5 to 16842518.6 oocytes. Due to the long reproductive period, recommendations include catch quotas, a closed season (February, March and June) and an 18 cm minimum size.

Keywords: reef fish; spawning frequency; fecundity; conservation.

Aspectos reproductivos de *Acanthurus chirurgus* (Perciformes: Acanthuridae) capturado por la flota artesanal que opera en la Isla de Itamaracá, Pernambuco, Brasil

Resumen: Este estudio examinó la biología reproductiva de *Acanthurus chirurgus* mediante muestreos mensuales desde mayo de 2016 hasta mayo de 2018. Se analizaron un total de 505 ejemplares (317 machos, 188 hembras), con los machos presentando una LT mayor que las hembras (32.0 cm vs. 30.8 cm). La mayor frecuencia de individuos se encontró en el rango de tamaño de 23–25 cm. La reproducción ocurrió de enero a julio, con intensidade reproductiva en febrero, marzo y junio. Las tallas de primera (L_{50}) y máxima maduración (L_{99}) fueron 18.0 cm y 22.1 cm, respectivamente. El desove fue sincrónico, ocurriendo en lotes y siguiendo un patrón de fecundidad indeterminado. La frecuencia de desove se estimó en cada 4.2 días (método HO) y cada 3.5 días (método POFs). La fecundidad por lote varió de 15.403,9 a 48.030,9 ovocitos, y la fecundidad relativa por lote de 2.710.628,5 a 16.842.518,6 ovocitos. Debido al prolongado período reproductivo, se recomiendan cuotas de captura, una veda (febrero, marzo, junio) y un tamaño mínimo de 18 cm.

Palabras clave: peces arrecifales; frecuencia de desove; fecundidade; conservación.

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INTRODUCTION

Acanthurus chirurgus (Bloch 1787) is a surgeonfish species found throughout the Brazilian reef bioprovince, mainly in coastal regions, from the continental shelf of the state of Maranhão to the coast of Santa Catarina (Ferreira et al. 2004). On the northeast coast, the species is abundant and is captured by the artisanal fleet operating with bottom traps (Figueiredo and Menezes 2000).

With the advancement of artisanal fishing, some reef fish species previously classified as bycatch have become valued in both domestic and international markets (Ribeiro 2006). Exports are primarily destined for the United States, Canada and some European countries. The United States is the main importer, accounting for 82.1% of exports (Carvalho et al. 2013).

Due to the high demand for fish consumption and the increased exploitation of traditionally harvested resources, *A. chirurgus*, which was formerly discarded, has become the target of specific fisheries (Marques and Ferreira 2010). On the northeast coast, the species is frequently captured by the artisanal fleet in the states of Ceará (Freitas et al. 2009) and Rio Grande do Norte, where it accounts for for 8.6% of the catches (Feitosa et al. 2002).

Given the ecological and economic importance of *A. chirurgus,* Rocha et al. (2002) evaluated habitat preference, larval dispersal and comparative phylogeography in Brazil, Ascension Island and Panama. Polunin and Roberts (1993) assessed the biomass of *A. chirurgus* in two marine reserves in the Caribbean. Robertson et al. (1993) analysed nesting and recruitment patterns from the Caribbean to Panama. Reeson (1983) studied the species' biology and ecology in the Caribbean.

Despite the studies described above, there is a lack of research focusing on the population structure and reproductive biology of *A. chirurgus.* This study aims to contribute to the understanding of the species' life history, particularly its reproductive aspects, providing relevant information to inform potential management measures that will aid in the conservation and sustainability of this fish stock. The information generated in this study includes previously unpublished data for Brazil.

This study aims to investigate the reproductive aspects of *A. chirurgus* in northeastern Brazil based on macro- and microscopic analyses of ovaries and testes. This includes a monthly evaluation of the gonadosomatic index, seasonal variation in maturation stages, spawning period, first maturation size, spawning frequency, spawning type and fecundity.

MATERIAL AND METHODS

Area of study and sampling

Fish were collected monthly from May 2016 to May 2018 at the artisanal fleet landings on Itamaracá Island, northern Pernambuco, Brazil (07°44.53'S, 34°49.49'W; Fig. 1). Specimens were caught using rectangular wooden pots with high-density polyethylene netting (98×95 cm, 40 cm diameter), deployed 20 nautical miles offshore at depths of 20-40 metres.

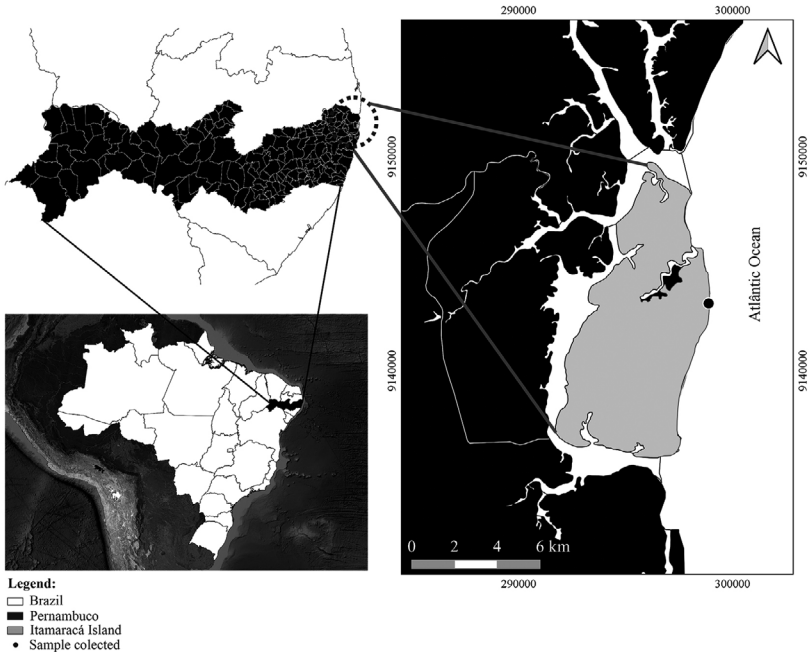

Fig. 1. – Location of the fishing area where *Acanthurus chirurgus* specimens were collected, northern coast of Pernambuco, Brazil (2016–2018).

For each specimen, total length (TL, cm), total weight (TW, g) and sex were recorded. Ovaries and testes were removed, weighed with a precision scale (0.01 g), fixed in 10% formaldehyde for 24 hours, and preserved in 70% alcohol. A ~1 cm cross-section of each sample was embedded in resin, sectioned at 5 μm with a LEICA RM2255 microtome, and stained with hematoxylin-eosin.

Maturation stages and spawning type

Histological analysis and imaging were conducted with an optical microscope (LEICA DM 500). Maturation stages were classified following Brown-Peterson et al. (2011), adapted for this species. Immature individuals that were not part of the reproductively active population were excluded from the gonadal maturation frequency analysis.

The spawning type of *A. chirurgus* was determined by measuring oocyte diameters in histological slides (n=200), using the largest diameter from the nucleus in samples at different ovarian development stages. Following Wallace and Selman (1981) and Murua et al. (2003), spawning types were classified based on oocyte development patterns.

Post-ovulatory follicles (POFs) were identified in ovarian histological sections as per Ferreri et al. (2021). The monthly spawning fraction was determined by the frequency of females with hydrated oocytes (HOs) and POFs, following Hunter and Macewicz (1985), estimating the proportion of females that are either about to spawn or have spawned in the last 12-24 hours.

Data analyses

Growth type and sex ratio

Descriptive statistics were used to calculate the mean and standard deviation of TL and TW by month and size class for each sex. Normality was tested with the Lilliefors test, variance homoscedasticity with the Bartlett test, and significant differences with the Shapiro-Wilk test (Zar 2010). Given homogeneous variances, ANOVA was used to compare means, followed by Tukey's post-hoc test at $p<0.05$.

The Mann-Whitney test was used to assess statistical differences in length classes between sexes. The sex ratio (males to females) was calculated monthly and analysed with the chi-square (χ^2) test to evaluate deviations from a 1:1 ratio. A significance level of $p=0.05$ was applied to all analyses.

Gonadosomatic index (GSI)

The reproductive cycle was assessed by analysing the relative frequency of ovarian (n=186) and testicular (n=293) maturation stages, along with the monthly variation in the gonadosomatic index (GSI). The GSI, calculated following Zudaire et al. (2013), was defined as follows: $GSI=(GW / EW)\times10^2$, where GW=gonad weight (g) and EW=eviscerated weight (g).

Environmental data

Principal component analysis (PCA) was used to examine relationships between rainfall, water temperature, months and development stages. Rainfall (mm) and water temperature (°C) data were sourced from the Pernambuco Water and Climate Agency (APAC) website, covering the monthly mean precipitation and water temperature for the region over the past 10 years.

Microscopic size at maturity (L_{50} and L_{99})

The minimum (L_{50}) and maximum (L_{99}) lengths of gonadal maturation were estimated using the logistic model $Mf=1/(1+e^{\beta2+\beta3TL})$, where Mf represents the fraction of mature individuals capable of reproduction, and β2 and β3 are the equation parameters (Mendes 1999). Statistical differences between the microscopic maturity of females and males were assessed using the Student t-test with a significance level of $p<0.05$.

Spawning frequency

Spawning frequency was determined by dividing the total number of mature females (100%) by the percentage of females with HO and POF in the ovaries. The ANOVA test was used to analyse statistically significant differences ($p<0.05$) between the frequencies of females with HO and POF.

The number of spawning events per year was estimated by multiplying the average spawning frequency by the length of the spawning season, a method commonly used in fish reproductive studies (e.g. Hunter and Macewicz 1985). The spawning season duration was based on the percentage of females with HOs in the ovaries throughout the period, following Hunter and Macewicz (1985).

Batch fecundity and relative batch fecundity

Batch fecundity (BF), defined as the number of oocytes in the germinal vesicle migration, germinal vesicle breakdown and HO phases that could be released per batch, was estimated for 15 spawning-capable females using three ovary subsamples (0.03±0.01 g) were collected from each ovary with the gravimetric method by counting the oocytes according to Murua et al. (2003). Each subsample was saturated with glycerin, and oocytes were counted under a stereomicroscope (Schaefer 2001).

BF was calculated using the formula BF=((ΣHi/Wi)/n) * OW, where Hi=number of oocytes in the germinal vesicle migration, germinal vesicle breakdown and HO phases for each repetition, Wi=weight of each ovary portion, n=number of repetitions, and OW=TW of each ovary.

Relative batch fecundity (BFrel) was estimated by dividing the BF by the gonad-free weight of the fish, estimated using the formula BFrel=(BF/(TW-OW)), where BFrel=relative batch fecundity, TW=total weight of each female, OW=ovary weight.

The relationships of BF and BFrel with TL, TW and OW in females were assessed using regression models. These models were further evaluated through a Z-test at a significance level of $p<0.05$ to determine the strength and significance of each predictor (Zar 2010).

RESULTS

Morphometry and sex ratio

A total of 505 *A. chirurgus* specimens were captured, including 315 males (62.37%) and 190 females (37.26%). Males ranged in TL from 11.9 to 32.0 cm (24.1±3.6). Females ranged from 12.9 to 30.8 cm (23.3±3.4). There were statistically significant differences between the average lengths of males and females (t=–2.4; df, 240; p=0.017), indicating that females were more likely to be smaller than males.

The highest frequency of TL for both sexes occurred in the 23–25 cm size class (Fig. 2). Statistical analysis showed no significant differences in length class distribution between sexes (Mann-Whitney U=46.50; p=0.35), with males predominating in most size classes.

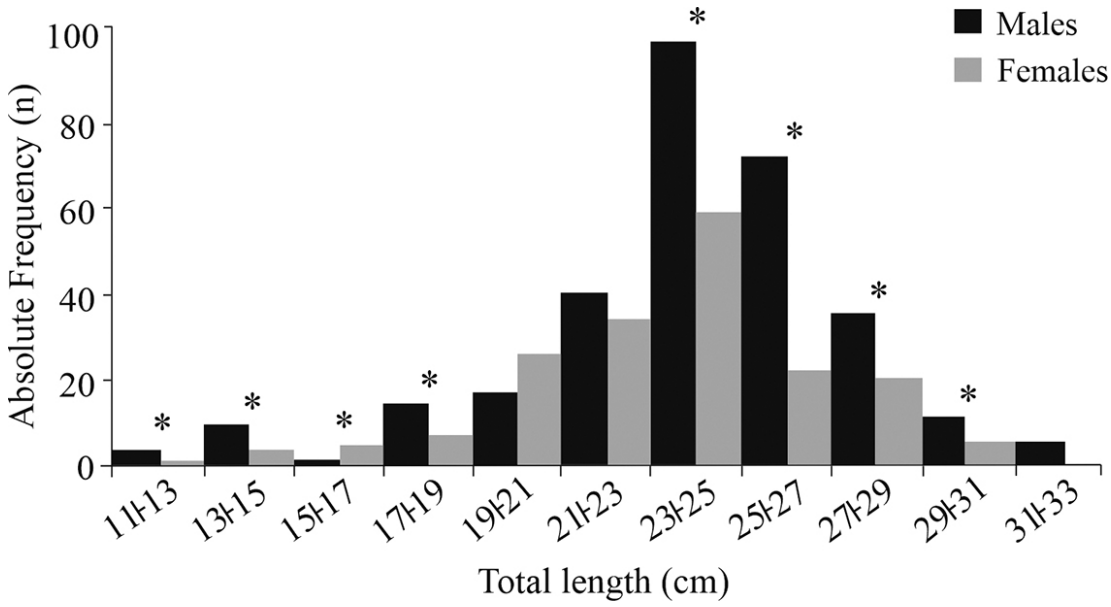

Fig. 2. – Comparative distribution of absolute frequency by length classes for male and female *A. chirurgus*. Statistically significant differences were found (Mann-Whitney test, $p<0.05$).

The estimated sex ratio for the sample period was 1.6 males to 1 female (1.6M:1F), significantly differing from the expected 1:1 ratio ($\chi^2=6.10$; df=1; p=0.01). Males predominated in most months, except in April, October and December (Table 1).

Table 1. – Monthly distribution, sex ratio, and χ^2 test for *A. chirurgus* captured on the northern coast of Pernambuco, Brazil, between 2016 and 2018. *Statistical differences.

Month	Males	Females	Total	M: F	χ^2 0.05;1=3.85
Jan.	27	13	40	2.1:1	12.3*
Feb.	36	13	49	2.8:1	22.0*
Mar.	22	10	32	2.2:1	14.1*
Apr.	10	17	27	0.6:1	6.7*
May	29	19	48	1.5:1	4.3*
Jun.	28	12	40	2.3:1	16.0*
Jul.	28	14	42	2.0:1	11.1*
Aug.	30	23	53	1.3:1	1.7
Sep.	41	17	58	2.4:1	17.1*
Oct.	15	15	30	1:1	-
Nov.	28	14	42	2.0:1	11.1*
Dec.	21	23	44	0.9:1	0.2
Total	315	190	505	1.6:1	6.1*

Gonadossomatic index

The monthly variation in the average gonadosomatic index (GSI) for females (Fig. 3) suggests that the reproductive period occurs from January to July, with spawning peaks during the months of February (1.66±0.71), March (1.78±0.84) and June (1.47±0.69), when the highest average GSI values were recorded, followed by falls. During this period, the weight of the gonads represented on average 0.55%, 1.35% and 0.63% of the gutted weight. From August to December, the GSI showed the lowest mean values, with fluctuations of between 0.39 and 0.66. From January, GSI increased (1.44±0.69), marking the beginning of the reproductive period. For males, small variations in average GSI values were recorded throughout the year. However, GSI increases were observed during the reproductive period (from January to June), with the highest average values being recorded in March and April (0.19±0.2), May (0.2±0.1) and June (0.21±0.2), while the lowest values occurred from July to December with fluctuations of between 0.06 and 0.09 (Fig. 3).

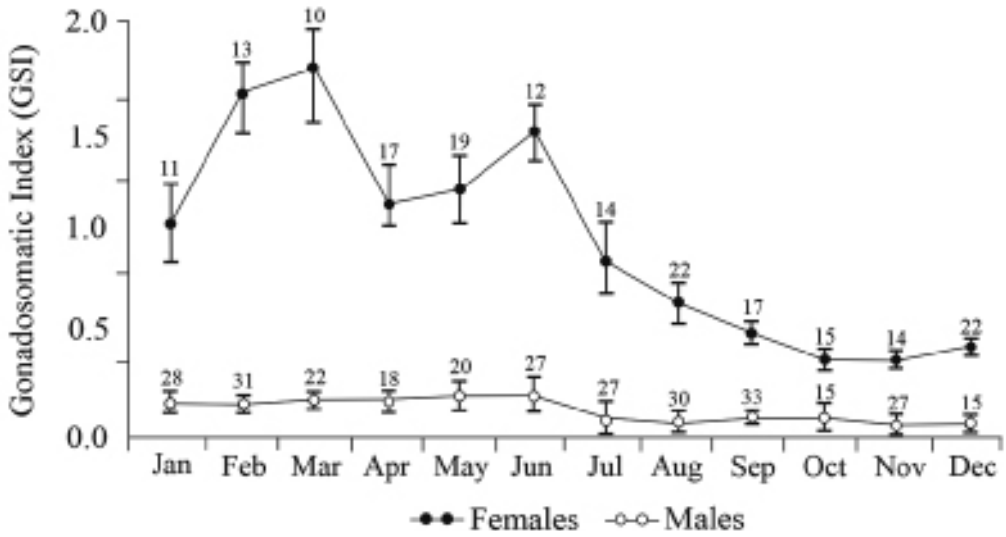

Fig. 3. – Monthly variation in GSI for female and male *A. chirurgus* captured on the northern coast of Pernambuco, Brazil (2016–2018).

Microscopic description of ovaries and testes

Microscopic analysis of the ovaries and testes enabled the classification of six development stages. Figures 4 and 5 summarizes the different oocyte and spermatozoa developmental stages, according to the classification described in Tables 2 and 3, respectively.

Females:

A) Immature: Presence of oogonia and oocytes in primary growth;

B) Developing: Primary growth present, with notable emergence of oocytes in cortical alveoli and primary vitellogenesis;
C) Spawning-capable: Includes all previous phases, with oocytes in secondary and tertiary vitellogenesis;
D) Actively spawning: Includes all previous phases, characterized by oocytes in germinal vesicle migration, germinal vesicle breakdown, HOs and POFs;
E) Regressing: Includes primary growth, but with a high abundance of oocyte atresia;
F) Regeneration: Features oocytes in primary growth, and in some cases, advanced atresia.

Males:
A) Immature: Formation of spermatocysts and presence of spermatogonia in the growth and proliferation zone. The lumen of the lobes is absent;
B) Developing: Spermatocysts present within the lobes, with spermatogonia, spermatocytes, spermatids, and a few spermatozoa. The lumen is filled with a non-cellular matrix;
C) Releasing-capable: All germ cell stages (spermatogonia, spermatocytes, spermatids and spermatozoa) are present, and the sperm ducts are filled with spermatozoa;
D) Releasing: Active spermatogenesis, with ongoing proliferation of spermatids and spermatozoa. The lumen of the seminiferous tubules is visible due to spermatozoa expulsion;
E) Regression: Residual spermatozoa are present in the lumen, marking the end of the active reproductive phase.

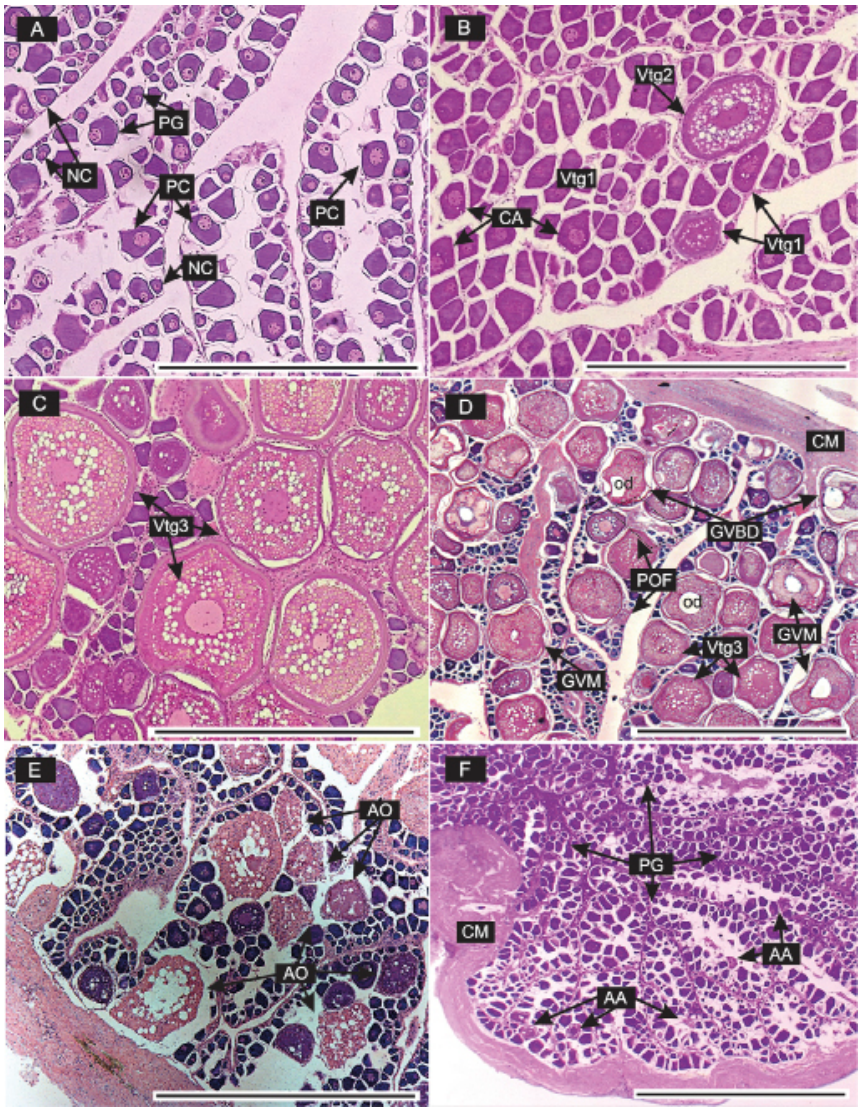

Fig. 4. – Ovaries of *A. chirurgus* at different maturational stages. Stages: immature (A), developing (B), spawning-capable (C), actively spawning (D), regression (E) and regeneration (F). Key: PG, primary growth; NC, nucleolar chromatin; PC, perinucleolar chromatin; CA, cortical alveolar; Vtg1–3, vitellogenesis stages; GVM, germinal vesicle migration; GVBD, germinal vesicle breakdown; Od, oil droplet; AO, atretic oocyte; AA, advanced atresia; CM, cell membrane. Staining: hematoxylin-eosin. Scale bar: 200 µm (A, B, C, D, E); 500 µm (F).

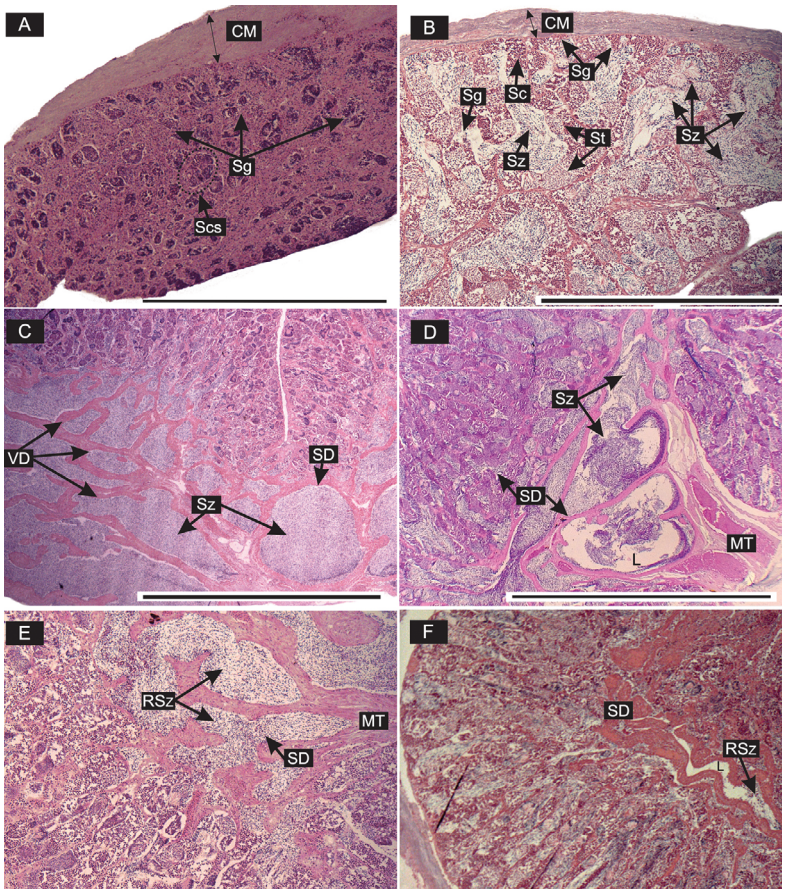

Fig. 5. – Testes of *A. chirurgus* at different maturational stages. Stages: immature (A), developing (B), releasing-capable (C), releasing (D), regression (E) and regeneration (F). Key: Sg, spermatogonia; Sc, spermatocyte; St, spermatid; Sz, spermatozoa; SCs, spermatocysts; RSz, residual spertmatozoa; L, lumen; VD, vas deferens; MT, muscle tissue; SD, sperm duct; CM, cell membrane. Staining: hematoxylin-eosin. Scale bar: 100 µm (A, B, E); 200 µm (C, F); 500 µm (D).

Table 2. – Summary of oocyte development stages in *A. chirurgus*. The morphological characteristics and sizes ranges are given for each oocyte stage. Measurements are made using histological slides.

Oocyte development stage	Characteristic	Cell diameter (µm)
Primary growth		
Chromatin nuclear	Large, centrally located nucleus is surrounded by a thin layer of cytoplasm and contains a single prominent nucleolus	27–59 x̄=46.0±7.3
Perinucleolar	Nucleus enlarges, with greater evidence of cytoplasm with multiple peripheral nucleoli	31–89 x̄=70.0±9.7
Cortical alveoli (CA)	Spherical vesicles appear in the cytoplasm. Oil droplets begin to accumulate, and the chorion and follicle layers become distinct	67–140 x̄=101.8±17.7
Vitellogenic (Vtg)	Appearance of yolk vesicles in the cytoplasm. It is further subdivided into three stage:	
Vtg1	Oil droplets occupy more cytoplasmic area than yolk granules	115–226 x̄=168.5±22.6
Vtg2	Oil droplets occupy similar cytoplasmic area than yolk granules	148–342 x̄=232.5±43.9
Vtg3	Oil droplets occupy less cytoplasmic area than yolk granules	201–446 x̄=327.5±51.2
Oocyte maturation (OM)		
Germinal Vesicle Migration (GVM)	Nucleus (germinal vesicle) starts to migrate to the animal pole and the oil droplets fuse to coalescence into a unique oil globule	288–577 x̄=426.1±48.2
Germinal Vesicle Breakdown (GVBD)	Nucleus completes its migration to the animal pole and the unique oil droplet is clear at central part of the oocyte	368–577 x̄=478.7±42.0
Hydration	Yolk granules merge into plates, creating a uniform mass, while the oocyte enlarges markedly from fluid uptake, becoming translucent	488–754 x̄=554.3±38.0

Table 3. – Summary of spermatozoa development stages in *A. chirurgus*. The morphological characteristics and sizes ranges are given for each oocyte stage. Measurements were made using histological slides.

Spermatozoa development stage	Characteristic	Cell diameter (µm)
Spermatogonia	These cells typically cluster at the spermatocyst periphery, with little cytoplasm and a prominent nucleus containing decondensed chromatin	3.0–5.4 $\bar{x}$=4.0 ±0.4
Spermatocytes	Cells cluster at spermatocyst edge, with little cytoplasm and prominent nucleus with decondensed chromatin	1.8–4.8 $\bar{x}$=3.0±0.7
Spermatids	Small cells with a reduced nucleus, condensed chromatin, minimal cytoplasm; marks end of spermatogenesis	1.5–3.3 $\bar{x}$=2.5 ±0.4
Spermatozoa	Flagellated cells with minimal cytoplasm; spermatozoa released into spermatocyst lumen and moved to spermatic ducts	1.5–3.2 $\bar{x}$=2.2 ±0.3

Environmental data and maturation stage

In the monthly distribution of maturity stages for females, the frequencies observed were 18.1% in the developing stage, 16.4% in the spawning-capable stage, 24.0% in the actively spawning stage, 25.2% in the regression stage and 16.4% in the regeneration stage (Fig. 6A). For males, the frequencies were 10.0% in the development stage, 16.1% in the releasing-capable stage, 20.4% in the releasing stage, 25.7% in the regression stage and 27.9% in the regeneration stage (Fig. 6B).

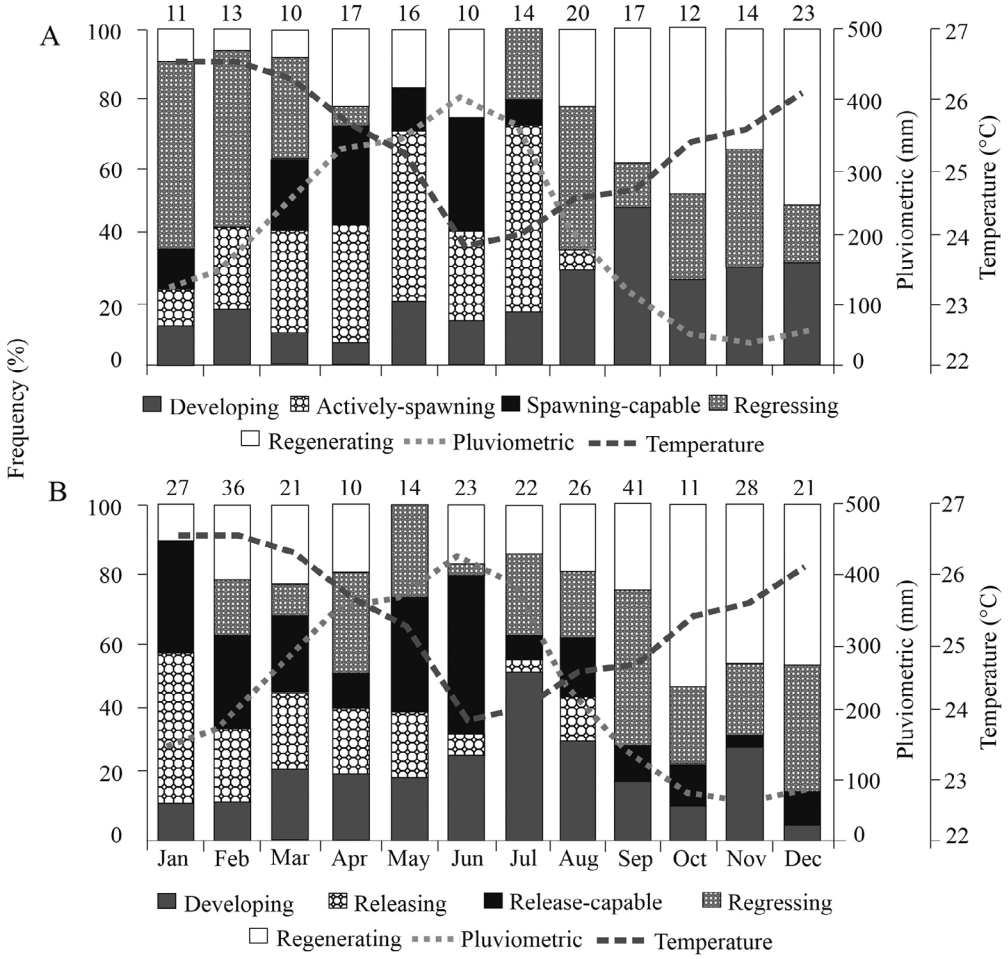

Fig. 6. – Monthly percentage distribution of gonadal maturation stages for females (A) and males (B) *A. chirurgus* correlated with rainfall (mm) and water temperature (°C) on the northern coast of Pernambuco, Brazil (2016–2018).

When the relative frequency of gonadal development stages was analysed, females were present in the developing stage throughout the year, with the highest frequency occurring in the second half, peaking in September (52.9%), October (25.0%), November (28.6%) and December (34.7%) (Fig. 6A). Mature females in the spawning-capable and actively spawning stages were observed from January to July, with the highest peaks in active spawning recorded in March (30.0%), April (35.0%), May (50.0%) and July (54.0%) (Fig. 6A).

The highest frequency of ovaries in regression was observed in August (35.0%) and November (35.7%). Conversely, the highest frequency of ovaries in regeneration occurred in September (51.2%), October (50%), November (37.7%) and December (52.2%), coinciding with the dry season characterized by average rainfall of 80.5 mm and water temperatures of 25.6°C (Fig. 6A). The presence of individuals with POFs and lower gonadal volume indicates the post-spawning season.

Males exhibited similar reproductive behaviour to females. Testes in the developing stage were observed by month, with the highest frequency occurring in the March and April (20%), July (36.4%) and November (30%) (Fig. 6B). Individuals in the releasing-capable stage were present throughout the sampling period, with the highest frequency being observed between January and June, particularly in January (44.4%), February (22.2%), March (23.8%), May (35.7%) and June (56.5%).

Individuals with mature testes were observed throughout the months. The highest frequency of males in the releasing stage was recorded from January to May, with notable peaks in January (33.3%), February (27.7%) and March (23.8%) (Fig. 6B).

Testes in the regression and regeneration stages were frequently observed throughout the sampling period, except for January (regression stage) and May (regeneration stage). The highest peaks for the regression stage occurred in September (43.9%) and December (38.1%). For the regeneration stage, the highest peaks were recorded in October (61.7%), November (45%) and December (47.6%) (Fig. 6B).

The reproductive cycle of *A. chirurgus* was classified as seasonal, with peak reproductive intensity in February, March and June. During this period, gonadal development was optimized, as evidenced by a higher frequency of females in the spawning-capable and actively spawning stages.

Analysis of rainfall patterns and water temperatures on the northern coast of Pernambuco from 2016 to 2018 revealed two distinct seasons: a dry season from August to February, with average rainfall of 90.5 mm and a water temperature of 25.2°C, and a rainy season from March to July, with average rainfall of 285.6 mm and a water temperature of 24.6°C (Fig. 6).

In the PCA, the first component explained 50.7% of the variance in maturity stages, while the second component explained 43.4%. Analysis of the extreme values along the PCA1 axis indicated a greater influence of months and rainfall, while the PCA2 axis showed a stronger influence of water temperature (Fig. 7).

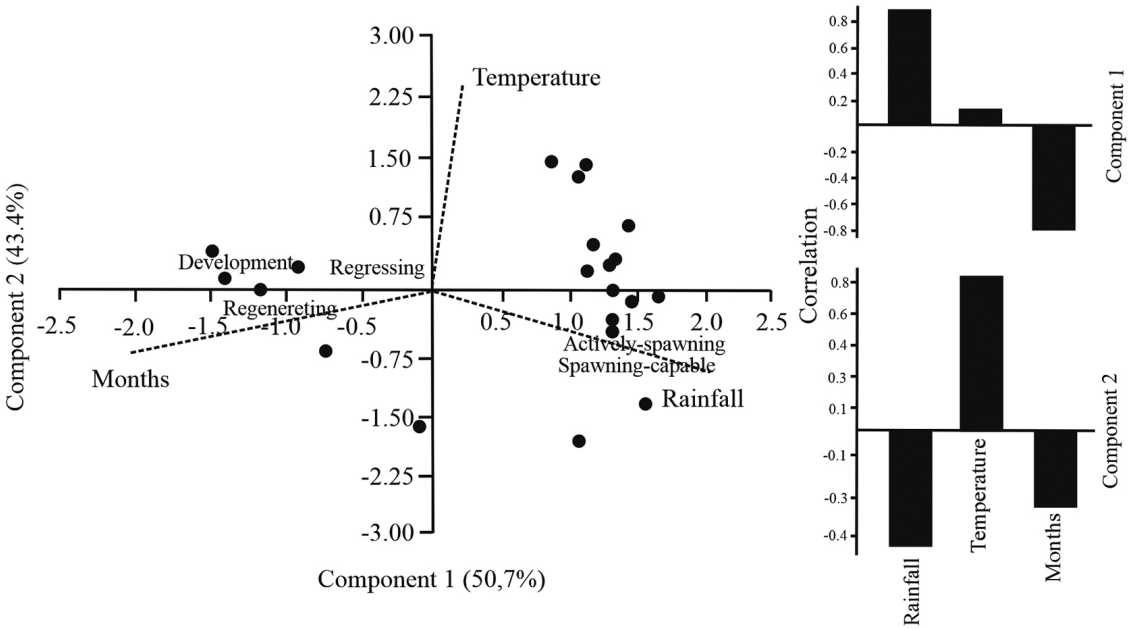

Fig. 7. – Principal component analysis (PCA) showing variance percentage and correlation with abiotic parameters for *A. chirurgus* captured on the northern coast of Pernambuco, Brazil (2016–2018).

These results align with the histological analysis of the ovaries and testes, which showed a gradual increase in developmental stages over several months. The PCA analysis supported the hypothesis that rainfall is directly related to the stages of development, as higher frequencies of spawning-capable and actively spawning stages were ob-

served between January and July (the rainy season). Conversely, the month variable had a greater influence on the development, regressing and regenerating stages, with contributions on axis 1 of 0.68 and -0.72, respectively. Water temperature contributed to distinguishing stages, with a value of 0.85 on axis 2, though it was not a dominant factor.

Maturation size

The comparison of maturity curves by length class for females and males revealed no significant statistical differences (p=0.64), suggesting that a single maturity curve can represent both sexes. According to the logistic model, the observed microscopic length at first maturation (L_{50}) for the species was 18.0 cm, with a confidence interval ranging from 17.7 to 18.7 cm (Table 4). The microscopic length at maximum maturation (L_{99}) was 22.0 cm, with confidence intervals between 21.3 and 22.5 cm (Fig. 8).

Table 4. – Logistic models of microscopic maturity for *A. chirurgus.* Key: R^2, coefficient of determination; L_{50}, size at first maturation; L_{99}, size at maximum maturation; MMF, microscopically mature females; MMM, microscopically mature males. Equal letters indicate statistical equality.

Microscopic maturity				
Equation		R^2	L_{50} (cm)	L_{99} (cm)
Females	$MMF = \frac{1}{1} + e^{[27.75+(-1.53TL)]}$	0.89	18.1	21.1[a]
Males	$MMM = \frac{1}{1} + e^{[27.95+(-1.53TL)]}$	0.89	18.2	21.1[a]
Both	$MM = \frac{1}{1} + e^{[27.32+(-1.74TL)]}$	0.91	18.0	22.1[b]

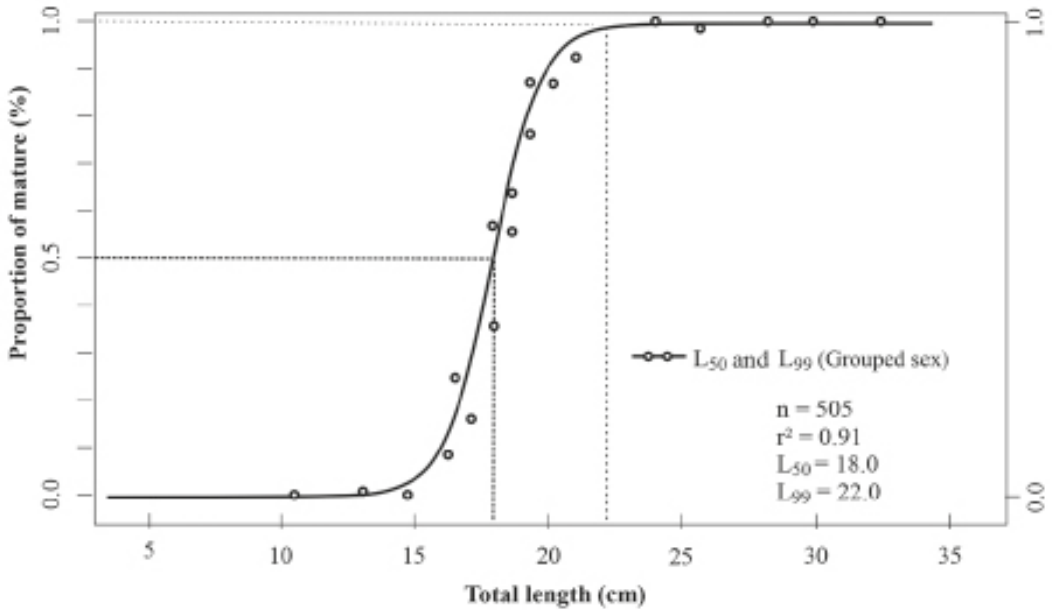

Fig. 8. – Maturation size (L_{50} and L_{99}) for *A. chirurgus* (both sexes combined).

Spawning type and spawning frequency

A. chirurgus exhibits batch spawning, is synchronous across multiple groups, and follows an indeterminate fecundity pattern. The size-frequency distribution of oocytes is multimodal (Fig. 9), with primary and secondary batches showing no gaps and diameters progressively increasing until spawning, as confirmed by histological analysis (Fig. 4). During the spawning season (January to July), 89 ovaries classified as spawning-capable and actively spawning were analysed. HOs and POFs were observed in 21 and 25 ovaries, respectively. The mean monthly spawning frequency was 23.4% (HOs method) and 28.3% (POFs method), indicating an average of 4.2 and 3.5 days between spawning events (Table 5). No significant difference was found between the methods (ANOVA, p=0.55), suggesting that each female spawns 29.8 times per year (HOs method) and 24.7 times (POFs method) over a 4.3-month spawning season.

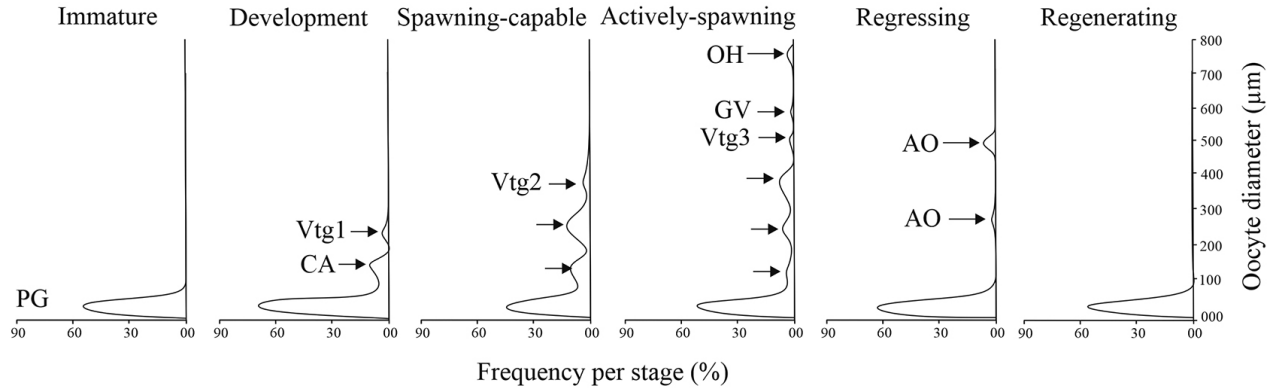

Fig. 9. – Frequency distribution of oocyte diameters across six ovarian development stages of *A. chirurgus.* (a) Visualization of all cell groups, with arrows indicating the formation of oocyte batches. Values represent the initial size of each class.

Table 5. – Spawning frequency of *A. chirurgus*. Key: N, total number mature ovaries; HO, number of ovaries with hydrated oocytes; POF, number of ovaries with post-ovulatory follicles.

Month	N	HO	POF	Monthly spawning (%)	
				HO	POF
Jan.	11	5	3	45.5	27.3
Feb.	13	3	4	23.11	30.8
Mar.	10	1	5	10	50
Apr.	15	3	1	20	6.7
May.	16	5	6	31.3	37.5
Jun.	10	2	1	20	10
Jul.	14	2	5	14.3	35.7
Mean	-	-	-	23.4	28.3

Batch fecundity and relative batch fecundity

BF ranged from 15403.9 to 48030.9 oocytes, with an average of 27102.1 (±11435.8) oocytes. The BFrel of *A. chirurgus* exhibited a wide variation, with values ranging from 2710628.47 to 16842518.6 oocytes per gram of body weight. The estimated mean was 8558391.9 (±4698 115.3), indicating a considerable dispersion of the data.

Figure 10 illustrates that both BF and BFrel of *Acanthurus chirurgus* increase with TL, OW and TW. The linear and potential relationships exhibited determination coefficients (R^2) ranging from 0.6487 to 0.7544 and 0.6364 to 0.6414, respectively, indicating significant correlations between these variables and fecundity ($p<0.05$).

The Z-test identified TW as the variable most strongly associated with BF (Z_1=2.2430, p=5.69×10^{-5}), suggesting that a percentage increase in TW results in an approximate 2.24-fold increase in fecundity. For BFrel, the Z-test indicated that OW is the strongest predictor (Z_1=2.7561, p=0.0012), implying that a percentage increase in OW leads to an approximately 2.75-fold increase in fecundity (Table 6).

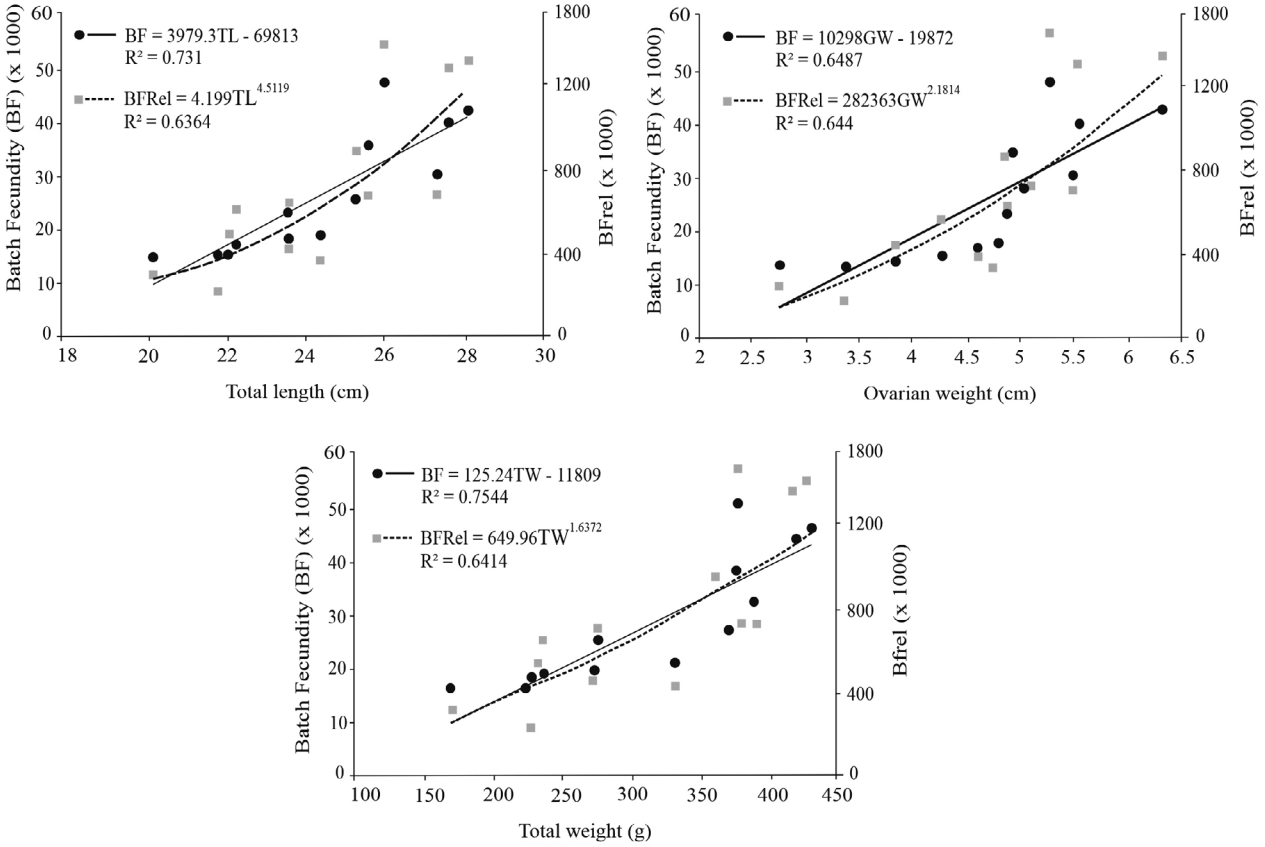

Fig. 10. – Relationship of batch fecundity (BF) and relative batch fecundity (BFRel) with total length, ovarian weight and total weight of mature female *A. chirurgus.*

Table 6. – Summary of regression models evaluating the relationship of batch fecundity and relative batch fecundity (batch fecundity/total weight) with total length, total weight and ovary weight in *A. chirurgus.*

Independent variable	Batch fecundity	Rel. batch fecundity
Model and Intercept (β_0)	Dependent variable; p-value	Dependent variable; p-value
$Log(BF)=\beta_0 + Z_1Log(TL)$	Z_1=1.3032; p=7.19*E* – 05	
$Log(BF/TL)=\beta_0 + Z_1Log(TL)$		Z_1=1.3032; p=0.0032
$Log(BF)=\beta_0 + Z_1Log(TW)$	Z_1=2.2430 (p=5.69*E* – 05)	
$Log(BF/TW)=\beta_0 + Z_1Log(TL)$		Z_1 2.2430; p=0.0049
$Log(BF)=\beta_0 + Z_1Log(OW)$	Z_1=0.4409 (p=0.0004)	
$Log(BF/OW)=\beta_0 + Z_1Log(OW)$		Z_1=2.7561; p=0.0012
Most significant predictor	Total weight (Z_1=2.2430)	Ovary weight (Z_1=2.7561)

DISCUSSION

The observed length difference between males (32.0 cm) and females (30.8 cm) of *A. chirurgus* suggests the presence of sexual size dimorphism, with males being slightly larger. This finding aligns with patterns commonly observed in many fish species, where size differences between sexes are often linked to specific reproductive strategies. Sexual size dimorphism can be an adaptive trait, with males potentially benefiting from increased body size for mate competition, while females may invest more energy in gamete production, as described by Bonduriansky et al. (2008). These differences in energy allocation can lead to distinct life history strategies that optimize reproductive success for both sexes.

The sex ratio of *A. chirurgus* (1.6M:1F) showed significant variations during the sampling period, likely due to reproductive strategies in which males compete for females. This behaviour may be associated with spawning

aggregations, as described by Hartup et al. (2013). The higher proportion of males in *A. chirurgus* may result from similar behaviours, particularly as fishing takes place on coral reef banks.

Differences in the sex ratio for some species may be associated with environmental conditions such as rainfall (Mozo 2006) and water temperature (Miltra et al. 2023) having an impact on the amount of energy available for growth, foraging and reproduction, differentiated sexual behaviour, growth rate and longevity. The observed variation in the sex ratio within the same stock of *A. chirurgus* may be influenced by fishing activity, seasonality and segregation in shoaling at feeding and spawning sites.

The catch size patterns for both sexes are likely influenced by gear selectivity, which leads to a tendency to capture larger individuals. Fish size is a primary factor of interest to fishermen, and larger fish are often being more valuable. In this study, the highest catches occurred in the 23 to 25 cm length class for both males and females. This finding corroborates that of Liang et al. (2012), who described that fishing gear selects fish through various mechanisms, the primary one being selection based on body length.

Males of the studied species were larger than females, measuring 32.0 cm and 30.8 cm, respectively. This size difference may be due to females allocating more energy to ovary development, as observed in *Haemulon plumieri* along the Pernambuco coast by Shinozaki-Mendes et al. (2013). Additionally, males were observed to reach slightly greater lengths, suggesting possible sexual dimorphism in *A. chirurgus*.

The GSI serves as an indicator of reproductive maturation stages. Higher GSI values, associated with increased gonadal activity (spawning-capable and actively spawning), were observed during the first semester, coinciding with the rainy season (March to July; mean rainfall=285 mm, water temperature=24.6°C). In contrast, lower GSI values, indicative of regression and regeneration stages, occurred during the drier second semester (August to February; average rainfall=90.5 mm, water temperature=25.2°C).

Studies on the reproductive period of Acanthuridae are limited and indicate considerable variation in the annual spawning patterns among species (Bushnell et al. 2010). In the present study, *A. chirurgus* exhibited a seasonal reproductive cycle, with gonadal development and spawning peaks occurring primarily during the rainy season, particularly in females.

PCA analysis of maturation stages and abiotic factors (rainfall and water temperature) revealed significant differences, with rainfall and temperature explaining the high frequency of mature females from January to July. This was followed by females in regeneration stages from August to December, supporting the idea of intense spawning during the first semester and a post-spawning period in the second semester.

Consistent with previous studies, the gonadosomatic index and maturation stage distribution indicate that *A. chirurgus* follows a seasonal reproductive cycle, with peak gonadal development and a high frequency of mature females during the first semester, corresponding to the rainy season. Increased rainfall may enhance nutrient availability in the coastal environment, stimulating primary biomass growth (Rodrigues and Pardal 2015). This factor likely influences the species' reproductive strategy, as *A. chirurgus* capitalizes on favourable conditions (higher rainfall and lower water temperature) to maximize reproductive success.

PCA showed that gonadal development stages are strongly influenced by rainfall and months (PCA1) and temperature (PCA2). The higher frequency of ovaries in regression or regeneration (inactive) stages may be linked to increased water temperature, which rises from 24.2°C during the rainy period to 25.6°C in the dry period. Ribeiro and Moreira (2012) noted that rainfall, water temperature and salinity variations in coastal zones are critical factors during larval incubation and hatching.

The high frequency of regression and regenerating in *A. chirurgus* individuals, even after spawning, may be linked to seasonal variations in water temperature and food availability, which constrain energy allocation to reproduction (Souza et al. 2014). According to these authors, such ecological pressures can delay gonadal development and prolong regression, serving as an adaptive strategy to enhance survival and optimize reproductive efficiency during favourable periods.

The microscopic L_{50} and L_{99} for *A. chirurgus* were 18.0 cm and 22.1 cm, respectively, for both sexes combined. Reeson (1983) reported an L_{50} of 17.0–17.9 cm for females and 14.0–14.9 cm for males in Jamaica. This discrepancy may be due to sampling limitations, capture methods or errors in classification during macroscopic analyses.

Our study's L_{50} analysis suggests that artisanal fisheries primarily target the adult stock, with only 7.52% of captured specimens being immature. This selective harvesting supports population sustainability by focusing on individuals that have already reached sexual maturity. Additionally, *A. chirurgus* exhibited oocytes at various developmental stages, confirming a batch spawning strategy, as indicated by the presence of mature and HOs—a key characteristic of batch spawners.

The synchronous spawning of *A. chirurgus,* with multiple oocyte groups developing and being released at intervals, indicates indeterminate fecundity—a pattern consistent with the continuous oocyte size-frequency distribution and the ongoing recruitment of primary growth oocytes throughout the spawning season (Murua and Saborido-Rey 2003, Ganias 2013).

This reproductive strategy, characterized by indeterminate fecundity and batch spawning, is common among marine fishes and enhances reproductive success by maximizing gamete output over an extended period, increasing fertilization chances in dynamic environments (Ganias et al. 2025).

The humid tropical climate and low latitudes of the tropical Atlantic favour cyclic ovarian development in *A. chirurgus*, with females spawning multiple times during the season and exhibiting synchronous spawning with multiple oocyte groups. For males, testes regeneration after sperm release suggests non-continuous maturation, optimizing reproductive performance and ensuring stock renewal. Cortés and Aron (2011) noted that batch spawning is a characteristic observed in most teleosts living at mid-latitudes. Additionally, Yamahira (2004) explained that species with a long reproductive season and multiple spawning events benefit from advantages that maximize reproductive success.

Estimating potential annual spawning numbers is crucial for *A. chirurgus.* Females are reproductively active for extended periods, spawning multiple times with a frequency of 4.2 days (HOs method) and 3.5 days (POFs method). This species has an estimated 29.8 and 24.7 potential batch spawning events per year, respectively.

Thus, estimating BF annual spawning numbers is essential for *A. chirurgus.* Females spawn multiple times throughout the year, with frequencies of 4.2 days (HOs method) and 3.5 days (POFs method). As a result, the species is estimated to have 29.8 and 24.7 potential batch spawning events annually, respectively.

BF and BFrel in *A. chirurgus* varied widely, averaging 27102.12 and 8558391.9 oocytes per female, respectively. Both metrics correlated strongly with TL, OW and TW, with the strongest associations observed for OW and TW. This finding suggests that larger fish with heavier ovaries produce more oocytes, reflecting a greater energy investment in reproduction. Power equations indicate possible nonlinear fecundity growth. These variations may stem from biological and environmental factors influencing reproduction, as noted by Bonduriansky et al. (2008).

BF data for *A. chirurgus* is not available in the literature, and oocyte production varies considerably among Acanthuridae species. The BF values determined for *A. chirurgus* align with those reported in previous studies. Queiroz et al. (2018) documented fecundity ranging from 20000 to 55000 oocytes in *Acanthurus coeruleus,* while Bushell (2010) reported 24000 to 44000 oocytes in *Zebrasoma flavescens.*

The variability in BF for *A. chirurgus* can be attributed to differences in female size, body shape and oocyte diameter used in different studies. Additionally, Ganias (2013) explained that low BF values may be compensated for by species that exhibit high spawning frequency, a behaviour also observed in *A. chirurgus.*

This study clarified the reproductive season of *A. chirurgus,* revealing no spatial segregation by maturity stage in either sex, with 93% of the catch comprising mature individuals and the stock being heavily exploited by the artisanal fleet in the northeast region. Considering the prolonged reproductive period (January and June), management measures are recommended, including catch quotas for the first semester, a closed season in February, March and June, and a minimum capture size of 18.0 cm for both sexes.

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CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

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CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Paulo R.S. Almeida: Writing – original draft, Visualization, Validation, Methodology, Formal analysis, Data curation, Conceptualization. **Renata A. Shinozaki-Mendes:** Writing – review, Visualization, Methodology, Formal analysis, Conceptualization. **María Korta:** Writing – review, Methodology, Visualization, Formal analysis, Conceptualization. **Hilario Murua:** Writing – review, Visualization, Methodology, Formal analysis, Conceptualization. **Paulo G.V. Oliveira:** Writing – review, Funding acquisition, Formal analysis, Supervision, Conceptualization.

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