The reproductive biology of the rainbow runner, *Elagatis bipinnulata* (Quoy & Gaimard, 1825) caught in the São Pedro and São Paulo Archipelago.

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(With 9 Figures)

Abstract

The rainbow runner, *Elagatis bipinnulata*, which belongs to the Carangidae family, has a circumtropical distribution. It is found throughout the Atlantic Ocean, from Massachusetts (USA) to Bahia (Brazil). The reproductive biology of the rainbow runner was studied, using specimens captured off the São Pedro and São Paulo Archipelago. From July 1999 to November 2003, a total of 352 fishes were analysed (201 females and 151 males). Fork length (FL) was measured and specimens were gutted for gonads collection. In the laboratory, gonads length, width and weight were measured, and sexes identified macroscopically. Through histological analysis, five different maturation stages were identified for females: immature, maturing, mature, spent and resting. A predominance of maturing and mature females was observed from January to May. The highest gonad index (GI) values were also observed during this period, ranging from 7.7 to 55. Mean sexual maturity size (L_{50}) for females was estimated at 64.6 cm (FL). In the studied area the species exhibited total spawning, with two synchronous groups. Testicles were histologically very similar making them impossible to differentiate sexual maturation stages. Considerable variation was observed in the male gonads weight, hampering the assessment of size at maturity. However, GI values for males were also higher from January to May. These results suggest that the spawning period of the rainbow runner in the São Pedro and São Paulo Archipelago occurs during the first semester of the year.

Keywords: spawning, gonadal index, fecundity.

Biologia reprodutiva do peixe-rei, *Elagatis bipinnulata* (Carangidae), capturado no Arquipélago de São Pedro e São Paulo

Resumo

O peixe-rei, *Elagatis bipinnulata,* da família Carangidae, apresenta distribuição circumtropical, sendo encontrado no Oceano Atlântico Oeste, desde Massachusetts (EUA), até o sul da Bahia (Brasil). O presente trabalho forneceu informações sobre a biologia reprodutiva desta espécie capturada no Arquipélago de São Pedro e São Paulo (ASPSP). Foram analisados 352 indivíduos (201 fêmeas e 151 machos), amostrados entre julho de 1999 e novembro de 2003. Os exemplares tiveram o seu comprimento furcal (CF) mensurado, sendo, em seguida, eviscerados para a coleta das gônadas. Em laboratório, foram medidos o comprimento, a largura e o peso das gônadas, identificando-se macroscopicamente o sexo. Histologicamente foram identificados cinco estádios de maturação sexual distintos para as fêmeas: imaturo, em maturação, maduro, desovado e em repouso. Observou-se uma maior frequência de fêmeas em maturação e maduras nos meses de janeiro a maio. No mesmo período, foram observados os maiores valores no Índice Gonadal (IG), que variou de 7,7 a 55. O tamanho médio de primeira maturação sexual (L_{50}) para as fêmeas foi estimado em 64,6 cm de CF. Para a área de estudo, identificou-se um tipo de desova total, sincrônica em dois grupos. As características histológicas dos machos amostrados foram muito parecidas, impossibilitando a diferenciação dos estádios de maturação sexual. Observou-se uma grande variação no peso das gônadas dos indivíduos que se encontravam no mesmo intervalo de

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comprimento furcal, tornando impossível a avaliação do tamanho com o qual iniciavam suas atividades reprodutivas. Entretanto, da mesma forma que as fêmeas, o IG dos machos também apresentou os maiores valores no período de janeiro a maio. Os resultados obtidos sugerem que o período de desova da espécie no Arquipélago de São Pedro e São Paulo ocorre no primeiro semestre do ano.

Palavras-chave: desova, índice gonadal, fecundidade.

1. Introduction

In 1988, the longline fishery based in Natal, Rio Grande do Norte state, began to operate around the São Pedro and São Paulo Archipelago, mainly from December to April, due to the higher abundance of yellowfin tuna (*Thunnus albacares*), which was caught by trolling and handlines (Hazin, 1993). Other bony fish species were also frequently caught around the archipelago, such as the wahoo (*Acanthocybium solandri*), the flying fish (*Cypselurus cyanopterus*) and the rainbow runner (*Elagatis bipinnulata*).

The rainbow runner belongs to the Carangidae family, and has a circumtropical distribution, occurring in the Atlantic from Bermuda and Massachusetts (USA), southward to northeastern Brazil (FAO, 2002).

Although *E. bipinnulata* is highly prized for its commercial and sport-fishing value, there is little information about its biology and ecology, especially migratory movements and feeding, which are still unknown. In Brazil, studies of its biology have only recently been initiated by Oliveira et al. (1997) and Vaske et al. (2006).

E. bipinnulata is a pelagic fish, normally found near the surface, over reefs, or occasionally over the islands slope. Although different types of gear are used to catch the rainbow runner, especially trolling and handline, it is not a species that is very easy to capture. Partially for this reason, little has been published about the species life-cycle. The study of the reproductive cycle, type of spawning and size at first sexual maturity for the rainbow runner, is of fundamental importance to ensure the sustainability of its exploitation, particularly in a remote insular ecosystem, such as the São Pedro e São Paulo Archipelago.

The aim of the present study, therefore, was to provide information on the reproductive biology of the rainbow runner, caught in the São Pedro e São Paulo Archipelago, by the longline fleet based in Natal.

2. Material and Methods

A total of 352 specimens were analysed. They were caught off the São Pedro and São Paulo Archipelago, by fishing vessels operating with trolling and handline, from July 1999 to November 2003 (Figure 1). Fishing activity was carried out at either dusk or dawn, using flying fish (*Cypselurus cyanopterus*) as bait, which was caught in the same area. Sampled specimens were identified, measured the fork length (FL), and eviscerated for gonad collection. In the laboratory, gonads weight, length and width were recorded. Sex was also identified, observing the gonads macroscopically.

The difference in length frequency distributions for separated sexes was examined by the Kolmogorov-Smirnov test, with the significance level of 5% (Snedecor and Cochran, 1989).

The sex ratio was estimated from the total number of males and females caught throughout the sampling period. To assess if the differences in sex ratio were statistically significant, the chi-square test (χ^2) was used, with the significance level of 5% (Snedecor and Cochran, 1989).

To determine the sexual stage of females, macroscopic observations of the ovary characteristics, such as colour and vascularisation, the presence of oocytes, together with the information of its volume in relation to the celomatic cavity, were done. The reproductive stages in females were determined using the Vazzoler (1981) scale, which is based on the macroscopic gonad aspects as follows: Stage A = immature; B = maturing; C = mature; D = spawned; and E = resting. On the other hand, for males, testicles size, colour and vascularisation, as well as the presence of seminal fluid were used for the classification of the reproductive stages.

For a more accurate determination of maturity stages, histological analysis on the gonads of 197 females was done. Cross-sectional cuts were made through the middle portion of the sampled gonads, and then dehydrated in alcohol, at concentrations of 80%, 90%, and 100%, cleared in xylol, and embedded in paraffin blocks. Sections by microtome were cut (6 μm) and stained with Harris´s haematoxylin, followed by eosin counter stain (Behmer et al., 1976). The cuts were read with the aid of an optical microscope (100x), and measured using a micrometric ocular.

The equation proposed by Schaeffer and Orange (1956) was applied to 149 adult females and 151 adult males for the calculation of the gonad index (GI), as follows:

$$GI = GW \times 10^5 / FL^3 \qquad (1)$$

where: GI = gonad index; GW = gonad weight (g); FL = fork length (cm).

The relative frequency (%) of adult individuals in each FL class was used to estimate the mean size at first maturity (L_{50}). For this analysis, individuals in stages B, C, D and E were considered adults (Vazzoler, 1981, 1996). A logistic curve was fitted to the observed data by the use of the following model:

$$Mf = 1 / [1 + \exp (a + b * FL)] \qquad (2)$$

where: Mf = is the maturity fraction of mature females; FL = fork length (cm).
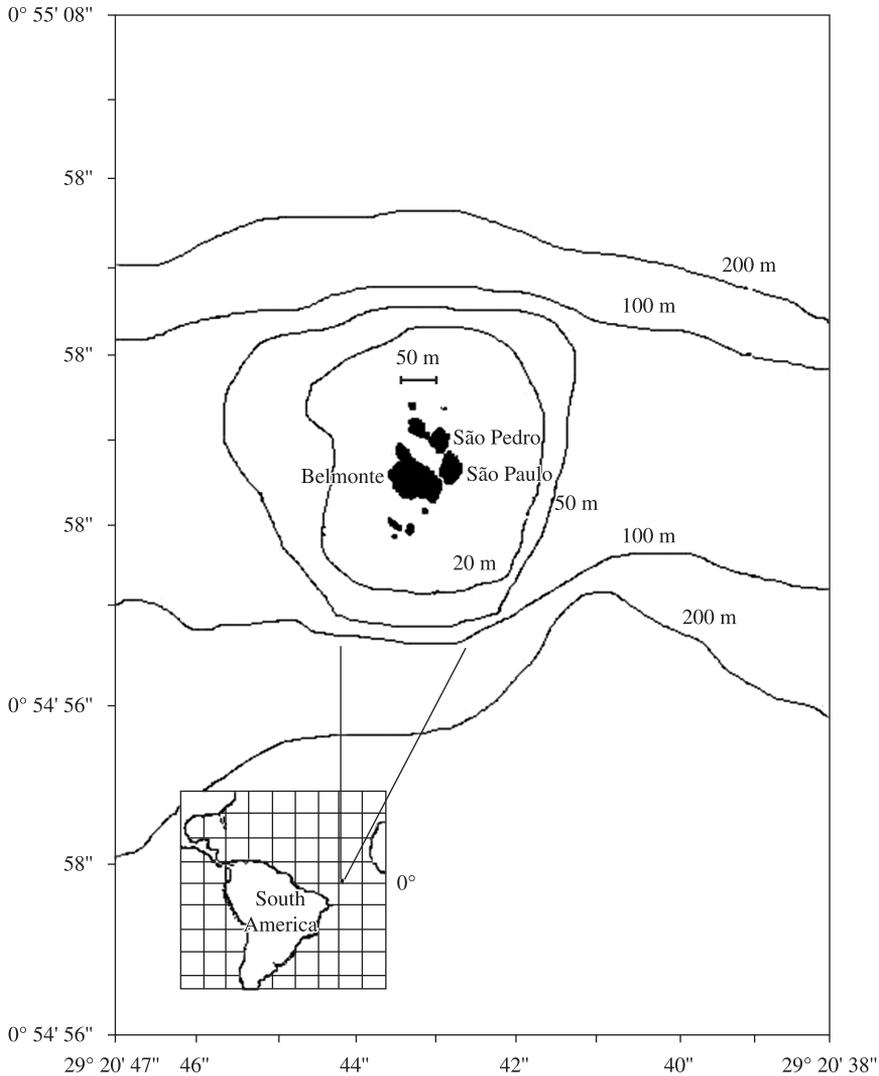
Figure 1. Study area: São Pedro and São Paulo (SS).

Fecundity was determined through the gravimetric method (Vazzoler, 1996). An aliquot of 0.05 to 0.08 g was removed from the middle portion of 15 sexually mature ovaries, and placed in a modified Gilson solution (Simpson, 1951) for the release of the oocytes, which were then counted and measured. The total number of oocytes in the ovaries (N) was estimated as follows:

$$N = n \times Wg / w \qquad (3)$$

where: w = the aliquot weight (g); n = the number of oocytes in the aliquot; Wg = the total weight of the ovaries (g).

The development of the frequency distributions of oocyte diameters and oocyte phases throughout different gonadal stages was used to determine the diameter at which oocytes complete their development before spawning. Oocytes with diameters greater than 0.4 mm were considered ready to spawn. Thus, the sum of the frequencies of oocytes ready to spawn (P) was used to estimate fecundity (F), as follows:

$$F = NP / 100 \qquad (4)$$

Spawning type was determined with the same 15 ovaries, by measuring 150 oocytes from each sample with the aid of a stereomicroscope equipped with a micrometer. The relative frequency (%) of oocytes per diameter class (mm) was then calculated.

Spawning season was determined from the frequency distribution of the different maturation stages and monthly variation of male and female gonad index (Vazzoler, 1996; Santos, 1978).

3. Results

Among the 352 individuals examined, 201 were female and 151 were male, resulting in a sex ratio of 1.3 females to 1.0 male. The female predominance was statistically significant ($\chi^2_{calculate} = 7.1 > \chi^2_{adjusted} = 3.8$, df = 1). Females were dominant most of the months, with the exception of

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March, April and May. However, differences in the monthly sex ratio were only significant (P = 1.3^{-10}) in August and September (Table 1).

The mean fork length (FL) for males was 70.5 cm (SD = 9.3 cm; n = 151) and ranged from 43 to 98 cm, but showed a bimodal distribution. According to Bhattacharya method (Sparre and Venema, 1998) the first mode occurring with an average of 62.2 cm in length with a standard deviation of 4.5 cm, and the second at 76.0 cm and standard deviation of 5.9 cm. The females had a mean FL of 69.8 cm (SD = 8.7 cm; n = 197) and ranged from 49 to 97 cm. Like males, females also showed a bimodal distribution, with the first mode in 60.4 cm and standard deviation of 5.4 cm, while the second trend was 74.6 cm and a standard deviation of 4.5 cm (Figure 2 and 3).

According to the Kolmogorov-Smirnov test, there was no statistically significant difference between the length compositions of males and females (calculated value = 0.11; tabulated value = 0.29).

Table 1. Monthly sex ratio of rainbow runner specimens captured around the S. Pedro e S. Paulo Archipelago during the period from July 1999 to November 2003.

Month	Females (n)	Males (n)	Total	Females (%)	Males (%)	χ^2
January	8	5	13	61.5	38.5	0.69
February	23	18	41	56.1	43.9	0.61
March	31	33	64	48.4	51.6	0.06
April	28	31	59	47.5	52.5	0.15
May	10	13	23	43.5	56.5	0.39
July	15	13	28	53.6	46.4	0.14
August	15	2	17	88.2	11.8	9.94*
September	37	19	56	66.1	33.9	5.79*
October	13	7	20	65.0	35.0	1.80
November	13	8	21	61.9	38.1	1.19
December	8	2	10	80.0	20.0	3.60
Total	201	151	352	57.1	42.9	7.10*

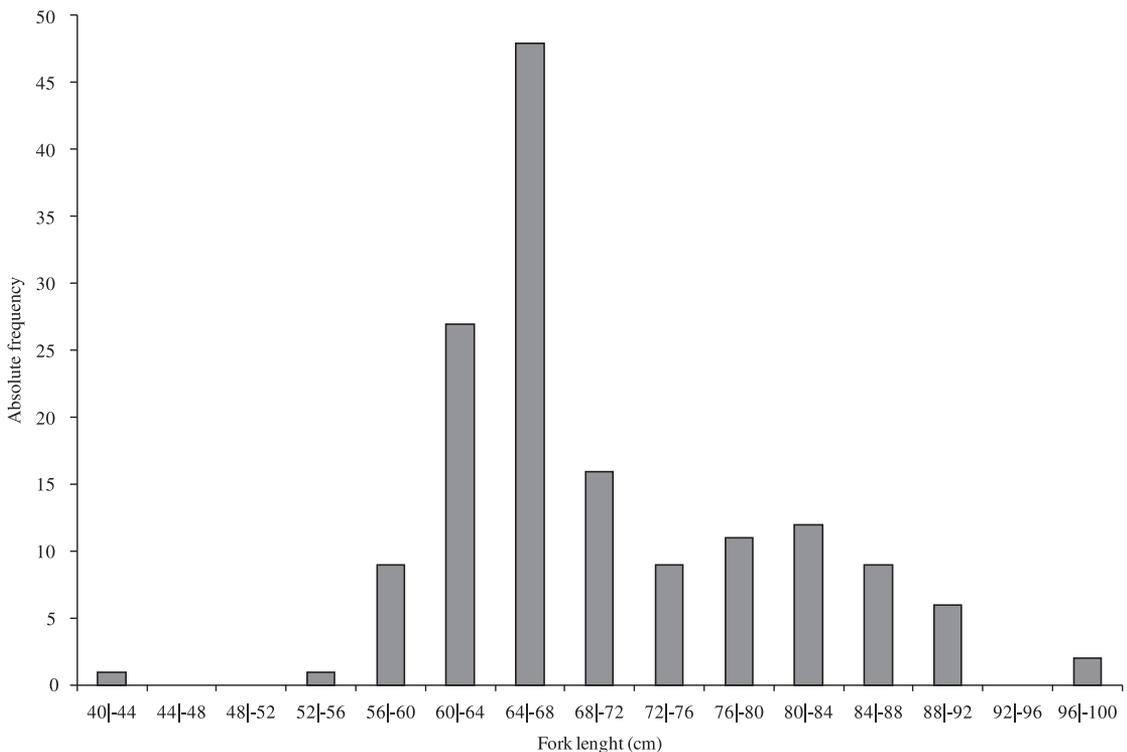*Significant difference at a level of 5%; p < 0.5.

Figure 2. Fork length frequency distributions of males rainbow runner, caught in the São Pedro e São Paulo Archipelago from July 1999 to November 2003 (μ = 70.5; σ = 9.3; n = 151).
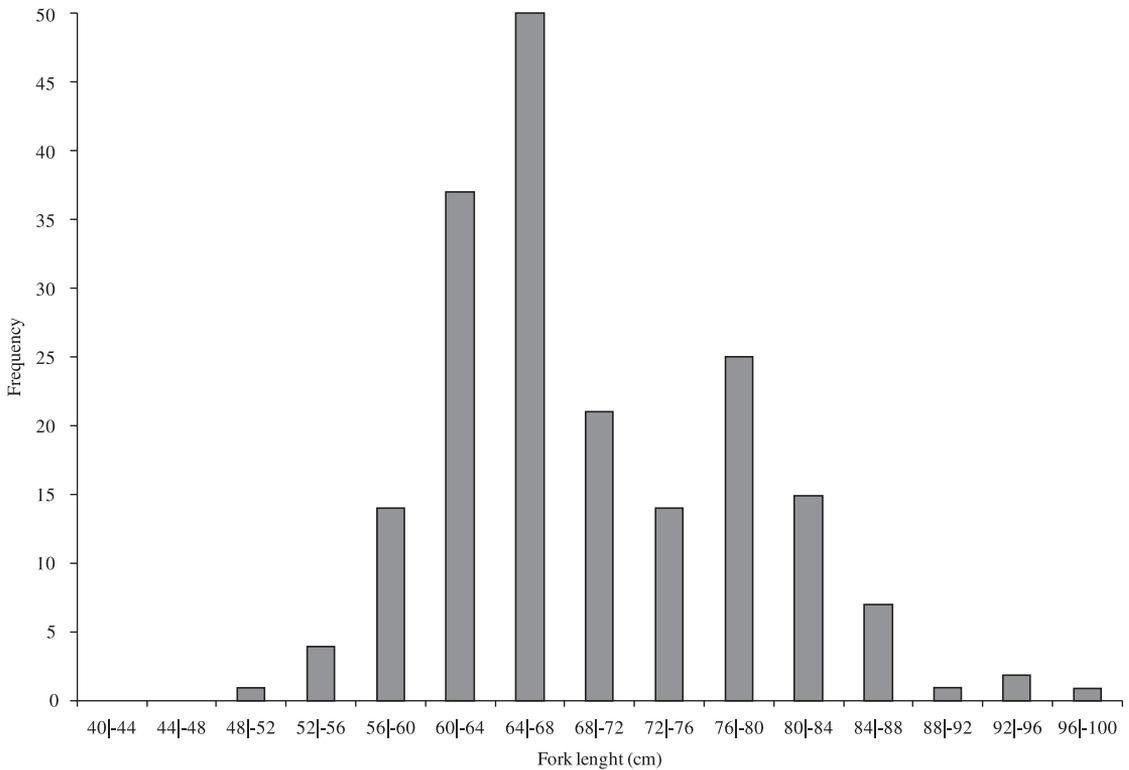
Figure 3. Fork length frequency distributions of females rainbow runner, caught in the São Pedro e São Paulo Archipelago from July 1999 to November (μ = 69.8; σ = 8.7; n = 201).

Higher frequencies of maturing and mature females were observed from January to July (no specimens were collected in june), whereas immature females predominated in August, September, November and December. Resting females occurred in March, April, July, September, but were particularly abundant from October to December (Figure 4).

From a total of 197 ovaries analysed histologically, 79 were maturing (40.1%), 44 individuals (22.3%) were mature and 43 (21.8%) were immature. Only two females had empty gonads (1.0%), whilst the remaining 29 females were resting (14.7%) (Figure 5).

Average GI for females ranged from 7.7 to 31.6, with the highest values (GI > 20) from January to May and in July, and the lowest values (GI < 20) in the remaining months (Figure 6).

The female mean size at first maturity, in the São Pedro and São Paulo Archipelago, estimated by the logistic model, was 64.6 cm FL (a = 12,79, p = 0.016 and b = -0,19, p = 0.013; r^2 = 0.90) (Figure 7).

Fecundity ranged from 1.376,400 to 2.627,680 oocytes, observed in specimens measuring 67.0 and 91.0 cm FL, with the gonads weighting 95.0 and 176.0 g respectively. Mean absolute fecundity was 2,024,876 oocytes (n = 15; σ = 471,670.3 oocytes). The relationship between absolute fecundity (F) and gonad weight (GW) was linear and positive (F = 9770.3 * GW + 688299.9; r = 0.91), whereas

between fecundity and fork length was linear and positive (F = 44416.9 * FL-1269377.5; r = 0.93) (Table 2).

The frequency distribution of oocyte diameter indicates the presence of two oocytes batches in distinct development stages. This pattern of ovarian development indicates that spawning is synchronous, with the oocytes ripening in two
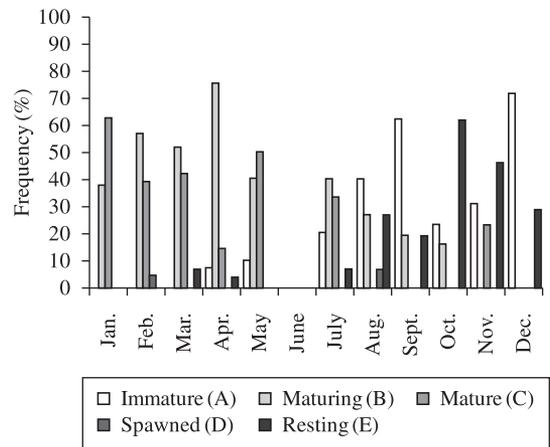

Figure 4. Monthly distribution of the sexual maturation stages of the rainbow runner females, caught in the São Pedro e São Paulo Archipelago from July 1999 to November 2003 (n = 197).
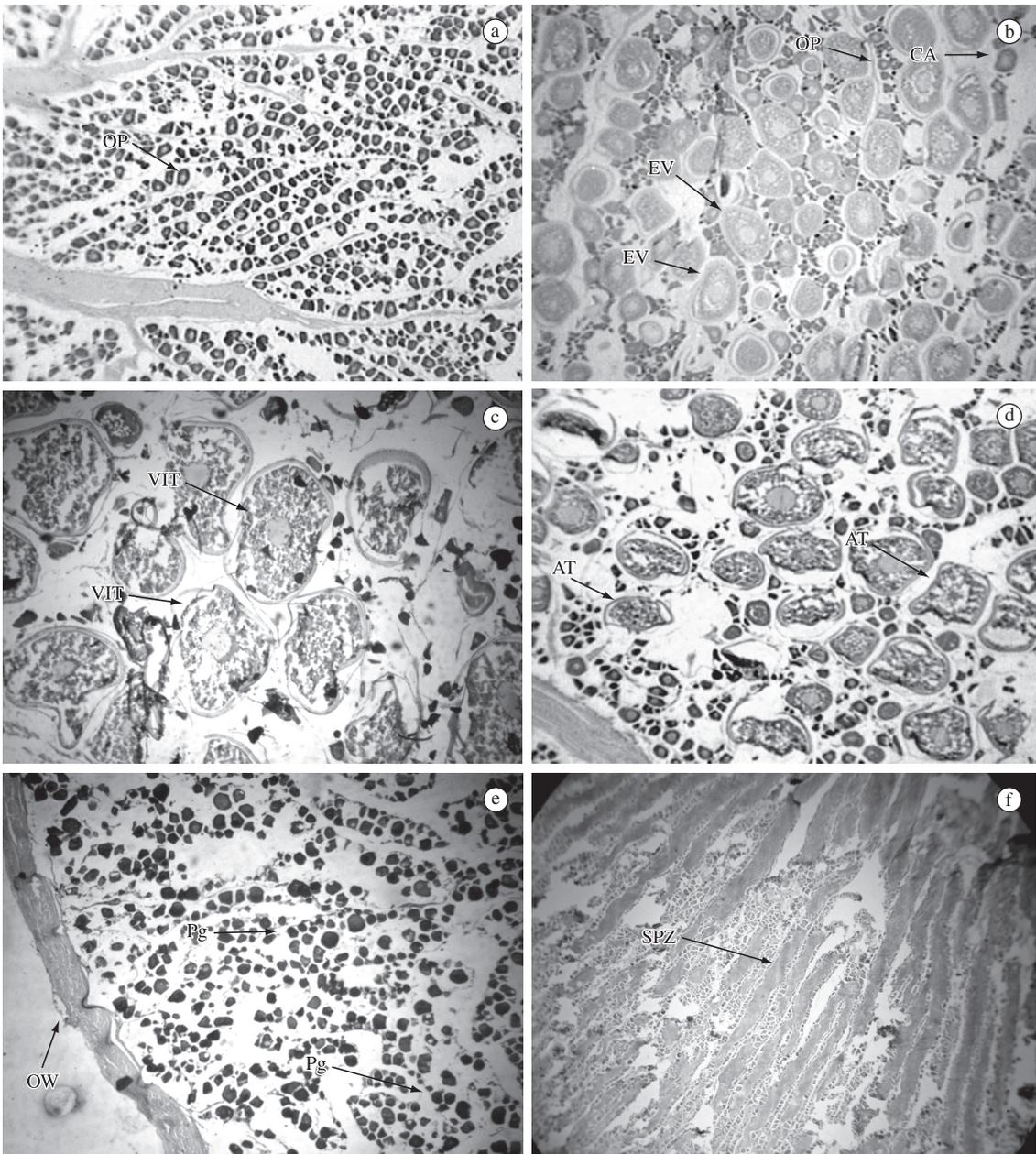
Figure 5. Histological sections in *E. bipinnulata* ovaries. a) Immature ovary; b) maturing ovary; c) mature ovary; d) spawned ovary; and e) resting ovary. (OP) perinucleolar oocyte; (EV) early vitelogenic oocytes; (CA) onset of cortical alveoli formation; (VIT) vitelogenic oocytes; (AT) atresia and folicules being absorbed; (Pg) previtellogenic primary growth oocytes; Ovarian wall (OW); and f) test – (SPZ) spermatozoa (100x; Haematoxilyn-Eosin).

groups. Therefore, it is characterised as a batch spawning. One oocyte batch was the reserve stock (R), while the second one was represented by the oocytes that would mature (M) (Figure 8). The modal class of the maturing oocytes ranged between 0.4 to 0.7 mm, suggesting that spawning probably happens around the larger diameter. Also the Bhattacharya method was used, in which there was a fashion at 0.4 mm, with standard deviation of 0.08, and the second mode at 0.6 mm and standard deviation of 0.06.

Male gonad weight ranged from 1.5 to 262.5 g. The relationship between gonad weight and fork length exhibited considerable dispersion; therefore it was impossible to determine the mean size at first maturity. It was also not possible to identify the maturation stages of males macroscopically in the sample. Although male gonads exhibited minor differences in size and volume, seminal fluid was observed in all the specimens. Similar characteristics were also observed in the histological analysis of the
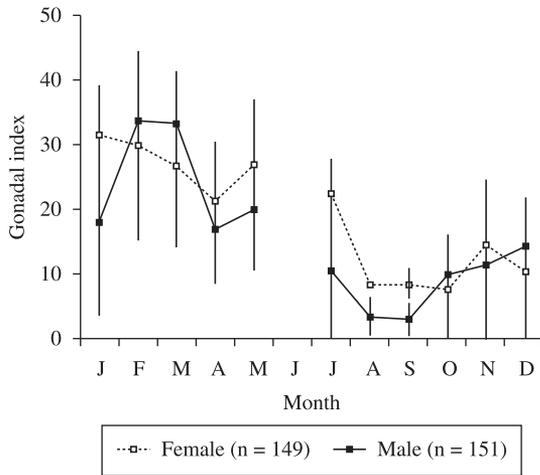
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Figure 6. Distribution of the mean gonad index by month for the rainbow runner males and females caught in the São Pedro e São Paulo Archipelago from July 1999 to November 2003 (n = 149). (Dark squares = mean, vertical bars = standard deviation).
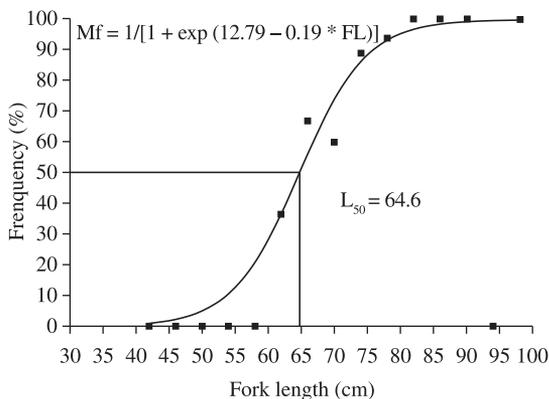

Figure 7. Logistic curve adjusted to the observed relationship between the relative frequency (%) of adult females and fork length (cm), for the rainbow runner caught in the São Pedro e São Paulo Archipelago from July 1999 to November 2003 (n = 103). The estimated mean size at first maturity (L_{50}) was 64.6 cm.

testicles, with the presence of spermatozoa in all cases (Figure 9). The monthly mean gonad index of males ranged from 3.1 to 33.8, with the highest values in February and March and the lowest in August and September (Figure 6).

The capture of *E. bipinnulata* ranged 401-6.222 kg in the period 1998 to 2003, observed two peaks of capture, one in 1999 (6.222 kg) and another in 2002 (5.787 kg), declining in subsequent years. A decline in fishing effort was also observed, starting in 2002 from 23.1 to 9.4 in 2003 (Figure 9).

4. Discussion

The rainbow runner in the São Pedro and São Paulo Archipelago exhibited a statistically significant difference
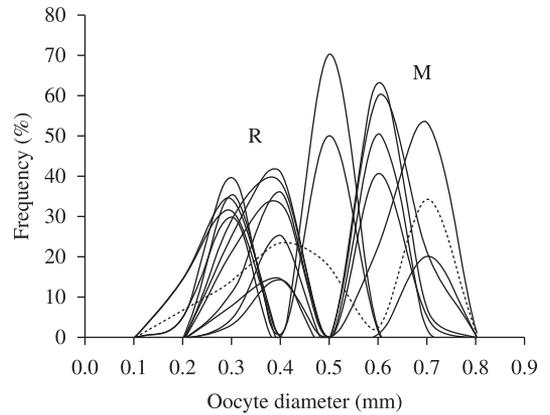

Figure 8. Relative frequency distributions of the female oocytes diameters (mm) for the rainbow runner, caught in the São Pedro e São Paulo Archipelago from July 1999 to November 2003 (n = 15). Dotted line = mean trend; R – reserve stock of oocytes; M – mature oocytes.
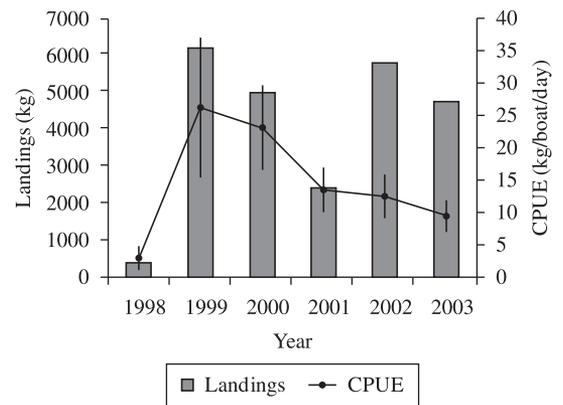

Figure 9. Annual distribution of catch per unit effort (CPUE) per kilo, boat and day of capture *Elagatis bipinnulata* caught in the São Pedro e São Paulo Archipelago from July 1998 to December 2003 (vertical bars indicate standard deviation).

in sex ratio (p = 0.0076), with an overall predominance of females (1.3F:1.0M), which might result from different mortality and growth rates between sexes. On the other hand, such differences might be simply due to sexual segregation, for reproductive reasons, which seems to be the present case, since higher proportions of males occurred between March and May, indicating a possible relationship between seasonal variations in sex ratio and the reproductive cycle, as both the gonad index and the frequency distribution of maturation stages suggest the first semester of the year as the spawning period for the species. Therefore, a sex ratio closer to 1:1 in this period could increase the possibility of a successful spawning.

The predominance of maturing and mature females from January to May, coinciding with higher gonad indices, seems to confirm a greater reproductive activity for the species in the studied area, during this period of the year. This characteristic was also observed in *Acantocibium solandri*, which likewise has a period of higher reproductive

Table 2. Model parameters of the regression the relationship between absolute fecundity (F) and gonad weight (GW) and between fecundity and fork length (FL), of rainbow runner specimens captured around the S. Pedro e S. Paulo Archipelago during the period from July 1999 to November 2003.

Relationship	R^2	Coefficient a	Significance (p)	Coefficient b	Significance (p)
F/GW	0.84	688299.9	0.00142	9770.3	1.7^{-6}
F/FL	0.86	-1269377.5	0.004	44416.9	5.8^{-7}

activity, in the same area, in the first half of the year (Viana, et al., in press).

Comparing the size at first sexual maturity for females (L_{50}; estimated at 64.6 cm FL) with the length frequency distribution of the specimens caught, it can be concluded that about 70% of females were above L_{50}, which is a positive aspect for the sustainability of the fisheries. Two modes were identified in the distribution of oocyte diameters, charactering a reserve stock and oocytes about to be released. This finding indicates total spawning synchronised in two groups. Vieira (2002) observed the same behaviour for the black fin tuna (*Thunnus atlanticus*) off the Rio Grande do Norte state (Brazil). Fishing for *Elagatis bipinnulata* held in ASPSP has been exploited in a sustainable way, since this species is not a target of the catch taken in the archipelago. Just as some species that occur in ASPSP, like *Acanthocybium solandri* (Viana, 2007), *E.bipinnulata* seems to reset its stock through a summer seasonal reproduction.

According to Viana et al. (2008), the most important fish caught (in weight) were tunas (55.7%), mainly yellowfin tuna, and wahoo (20.6%). Other species, including rainbow runner (*Elagatis bipinnulata*), black jack (*Caranx lugubris*), dolphin fish (*Coryphaena hippurus*), billfishes (*Istiophorus albicans, Makaira nigricans, Tetrapturus albidus*), swordfish (*Xiphias gladius*), made up 6.1% of the total catch. Fishing in the Archipelago is not directed to the *E. bipinnulata*, so the stocks of this species in the area are not under threat, since we observed a decrease in CPUE over the years.

According to Oliveira et al. (1997), from 1994 through 1996, 411 t of fish were caught around SPSPA. Of these, the flying-fish accounted for 41.7%, tunas for 30.6%. From 1998 onwards, therefore, there seemed to be a clear inversion of the predominant species, with the participation of flying-fish in the catches, by weight, dropping to only 13%, while the percentage of tunas increased to 56%. This reduction can be by changing the strategy of the fishing fleet, directing the catch for tuna, since its value commercial is superior to other species.

We hope that the results herein presented can contribute to a more efficient management of the fishing activity around the Archipelago, as well as to a better understanding of the reproductive biology of *E. bipinnulata* in the area, since information about the species is quite scarce.

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