Reproductive biology of the mullet *Mugil liza* (Teleostei: Mugilidae) in a tropical Brazilian bay

Rafael J. Albieri & Francisco G. Araújo

1 Laboratório de Ecologia de Peixes, Universidade Federal Rural do Rio de Janeiro. Antiga Rodovia Rio-São Paulo, km 47, 23851-970 Seropédica, Rio de Janeiro, Brazil.

2 Corresponding author. E-mail: gerson@ufrrj.br

**ABSTRACT.** The reproductive biology of *Mugil liza* Valenciennes, 1836 is described as a contribution to an elaborate management program. A total of 243 specimens (89 males and 154 females) were collected in the Sepetiba Bay in southeastern Brazil from July/2006 to June/2007. The gonadosomatic index ($I_G$) and the sequential development of the ovaries observed through histological studies suggested that the spawning season ranged from May to August. The population reached total sexual maturity ($L_{100}$) at 550 and 570 mm total length ($L_T$) for males and females, respectively. Females attained a larger size than males, and the sex ratio was female-biased for fish larger than 500 mm $L_T$. The hepatosomatic index ($I_H$) was significantly related to the $I_G$, indicating that vitellogenesis mobilizes hepatic energy during reproduction. Mean fecundity was 3,080,000 oocytes. The presence of only two phases of oocyte development in ripe ovaries – a reserve stock and a clutch of post-vitellogenic oocytes – indicated that ovarian development is group synchronous and this species is characterized as a total spawner. The results suggest that establishing a closed fishing season from May to August and establishing a minimum size for capture of 350 mm $L_T$ would enhance stock conservation and production for future harvest seasons.

**KEY WORDS.** Hepatosomatic index; oocyte development; spawning; stock conservation.

The mullet *Mugil liza* Valenciennes, 1836 is a coastal species distributed in the western Atlantic, from the Caribbean to Southeastern Brazil (Froese & Pauly 2008). Overall, mullets are reported to be semi-catadromous, with the juveniles being recruited to lagoons and estuaries following a period of offshore spawning (Ditty & Shaw 1996, Blaber 2000).

Mullets are important economic resources that support many small communities through both fishing and aquaculture (Pina & Chaves 2005, Katselis et al. 2005). The state of Rio de Janeiro is the most important producer of *M. liza* in southeastern Brazil, surpassing 1,000 ton x yr⁻¹, mainly from artisanal catches (IBAMA 2005). In spite of the importance of mullets to fishery resources in Rio de Janeiro, no management policies have been established for them in the Sepetiba bay, a large bay in southeastern portion of the state. One of the reasons for the latter is that wildlife managers require scientific research to take protective measures, such as imposing a “closed season.”

*Mugil liza* represented 99.2% of the total number of Mugilidae caught in experimental samples between 1994 and 1997 (Silva & Araújo 2000). In spite of its abundance, this species has been poorly studied, and little information is available on its life cycle in wild conditions (Alvarenga-Lajonchere 1979, 1981, Benetti & Netto 1991). To date, there have been no histological observations of seasonal variations in the development of the gonadal cells and the energy in the somatic tissues of *M. liza* from the Brazilian coast. The aim of this study is to describe the reproductive biology of *M. liza* from a coastal bay in southeastern Brazil. We test the hypothesis that the reproductive pattern of *M. liza* is similar to that of other mullets such as *Mugil platamun* Günther, 1880 (Andrade-Talmelli et al. 1996, Romagosa et al. 2000) and *Mugil cephalus* Linnaeus, 1758 (McDonough et al. 2003), by investigating key parameters such as gonadal development, fecundity and spawning season. Since *M. liza* is suffering a great fishing pressure in the Sepetiba Bay, we provide technical data that can contribute to the establishment of a management program.

**MATERIAL AND METHODS**

The Sepetiba Bay is located in the southeastern region of the Rio de Janeiro State (22°54'23"04'S, 43°34'44"10"W, Fig. 1) and has an area of ca. 450 km². The bay has two distinct zones. The inner zone, influenced by rivers and tidal creeks, has a predominantly muddy substratum and beaches that are rocky, sandy, muddy, and in places, fringed with mangrove formations. The outer zone is closer to the sea, and has a predominantly coarse and sandy substratum; rocky islands are common. About 40% of the bay area is less than 5 m deep; the maximum depth, up to...
were packed in ice and transported to the lab. Total length (LT) and Metereological Base.
collected at www.rio.rj.gov.br/georio from the Sepetiba and 55 mm between opposite knots). Data on the rainfall was 3 m high, and had three panels of different mesh sizes (45, 50, the inner zone of the Sepetiba Bay. The nets were 1500 m long, July 2006 to June 2007. MIFA uses small boats that operate in catches of the Madeira Island Fisheries Association (MIFA) from 23°00'S 22°45'S 05' 10' 55' 50' 44°00' 05' 10' 0 20 km 20 km 0 20 km Sepetiba Bay Atlantic Ocean REO J. Albieri & F. G. Araújo 30 m, is near to the sea limit. Salinity ranges from 28 (rainy season) to 34 (dry season), and the temperature varies from 21.5°C in winter to 27°C in summer (ARAÚJO et al. 2002). The tide amplitude is ca. 1 m, and southwestern and northeastern winds contribute to moving seawater into the bay and taking bay water out toward the continental shelf, respectively (SIGNORINI 1980).

The rainfall period in the bay region occurs mainly between December and January (summer), though it can sometimes last until March. The dry period extends from May to September (winter). South quadrant winds and marine breezes discharge their moisture against the mountain cliffs around September (winter). South quadrant winds and marine breezes can increase the amount of rain in the dry season (temporal (monthly) changes in the means of IG, IH, and K were tested with a Chi-square (x²) test. The relationship between proportion of mature fish (P) in each length interval was described by logistic model, P = 1/(1 + e-ª x LT - 1)), where, a and b are constants and can be estimated by procedure NLin based on 89 males and 154 females. Size at first maturity (L₅₀) and size when the whole population was mature (L₁₀₀) was then obtained by substituting P = 0.5 and P = 1, respectively, in the above equation.

The gonadosomatic index (IG = WG x WE x LT - 1 x 1000). The IG, IH, and K were tested for eventual correlation by using the Spearman test (rₛ). A Kruskal-Wallis non-parametric test was applied to determine whether the frequency distribution of gonad maturation stages determined the gonadal cycle. Fish condition was established through the hepatosomatic index (IH = WL x WE x LT - 1 x 100) and the condition factor (K = [WG x WE x LT - 1 x 100]. The IG, IH, and K were tested for significant differences (p < 0.05). Eviscerated weight was used in all calculations to avoid the influence of the contents of the gonad and stomach on the weights. All data are expressed as means ± standard error.

Fecundity was estimated by the gravimetric method and calculated as follows: F = N x WC x WGS - 1, where F = fecundity, N = number of post-vitellogenic oocytes from gonad sub-sample, WC = gonad weight and WGS = gonad sub-sample weight.

RESULTS

Gonads Morphology

Gonads are paired, elongated, covered by a thin peritoneal layer, and range from filiform to piriiform in ovaries or from a filiform to a thicker ribbon-like form in testes, depending on the developmental stage (Tab. 1). Cranial regions are larger, becoming thinner towards the caudal portion. Each gonad duct lies on the dorsal-medial region. These ducts have a small joint leading to a common orifice. Through the gonads, the arteries occupy a supra-visceral position and spread through lateral ramifications that become evident during gonad development. The right gonad is usually larger than the left.
Histological observations
Immature ovaries contained germ cells (and young oocytes) undergoing profound changes in their nuclear structure, cytoplasm, and membranes. The oocyte development was classified into two stages: the previtellogenic stage (germ cells, young oocytes, and peri-nucleolus oocytes from reserve stock) and the vitellogenic stage (oocytes with lipid vitellogenesis, oocytes with lipid and protein vitellogenesis, and oocytes at the post-vitellogenic stage) (Figs 2-7, Tab. II).

The testes are involved with the tunica albuginea and contain the seminiferous tubules. Internal to the seminiferous tubules are Sertoli cells that surround the cysts formed by spermatogenesis cells, which are all smaller than 10 µm (spermatogonia, primary and secondary spermatocytes, spermatids, and spermatozoa). Functionally, the testes were divided into four stages according to cellular type: immature, maturing, functional maturation, and recovering (Figs 8-13, Tab. II).

The size of M. liza ranged from 285 to 500 mm L_T for males (n = 89) and 325 to 690 mm L_T for females (n = 154). The number of females longer than 500 mm significantly outnumbered the number of males (size class 500-550 mm L_T: 1.0: 2.5; $\chi^2_{0.05} = 3.85$, d.f. = 1). The ratio for the entire sample (n = 243) was 1.0 male to 1.73 female ($\chi^2_{0.05} = 17.38$, d.f. = 1, Tab. III). The size at maturity (L_{T50}) for M. liza was 350 mm for females. The L_{T50} was 570 mm and 550 mm for females and males, respectively. For males, the L_{T50} was not calculated due to the small number of immature fishes.

The mean I_H also showed seasonal differences (H = 38.54, p = 0.0001, Fig. 14), with the lowest values between October and March (0.08 to 0.03 ± 0.003, respectively) and the highest values in June (2.09 ± 0.59), July (1.0 ± 0.01), and August (0.9 ± 0.01).

Ripe/running ripe ovaries were observed between May (25%) and August (12%). Spent ovaries were recorded between May (8.8%) and September (7.7%). Immature ovaries with germ cells were observed between July (25%) and February (46.6%); developing ovaries with yolk vesicle oocytes and recovering/resting ovaries with peri-nucleolus stage oocytes were found throughout the study period. Maturing ovaries with lipid protein vitellogenesis were observed between April (25%) and September (7.7%). Ripe and spent testes were not observed during the study, but maturing testes with spermatozoa were recorded mostly between March (18.2%) and June (53.8%) (Fig. 15).

The mean K and L showed seasonal differences during the study period for males (L_{K}: H = 34.02, p = 0.0004; K:H = 25.38, p = 0.0080) and females (L_{K}: H = 57.54, p < 0.0001; K:H = 27.30, p = 0.0041) (Fig. 14). From December (1.65 ± 0.07) onwards, the L_{K} for females gradually increased, reaching a peak in June (2.30 ± 0.12); the L_{K} then decreased from August (1.68 ± 0.13) to November (1.32 ± 0.06), when the lowest value was found. The L_{K} for males also increased from December onward, and reached a peak in February (2.48 ± 0.11). The L_{K} then decreased slightly to 2.00 ± 0.09 in July and dropped in the subsequent months to the lowest values in November (1.40 ± 0.06).

The mean K did not show a well-defined seasonal pattern of variation, and shifted throughout the study period. However, a trend was observed: values were low between June (8.50 ± 0.22) and November (8.31 ± 0.19), and high between December (9.16 ± 0.22) and May (8.93 ± 0.19) for females; males had low values between July (8.65 ± 0.13) and January (8.50 ± 0.13), and high values between February (9.65 ± 0.11) and June (8.92 ± 0.09). A significant positive relationship was detected...
Figures 2-7. Histological sections of ovary at different maturity stages of *Mugil liza* from Sepetiba Bay: (2) immature ovary containing germ cells and young oocytes; (3) ovary during the vitellogenesis process; (4) developing ovary containing lipid vitellogenesis oocytes; (5) maturing ovary containing lipid and protein vitellogenesis; (6) ripe/running ripe ovary containing post-vitellogenic oocytes; (7) recovering/resting ovary containing empty follicle. (gc) Germ cells, (yo) young oocytes, (rs) reserve stock – peri-nucleolus stage, (v) vitellogenic oocyte, (l) lamella, (pn) peri-nucleolus stage, (fc) follicular cells – forming follicular layer, (n) nucleus, (yg) yolk globule, (nc) nucleolus, (od) oil droplet, (pg) protein granules, (vm) vitelline membrane, (ao) atretic oocyte, (pvo) post-vitellogenic oocytes, (lv) lipid vesicles, (ef) empty follicle. Scale bars: (2) = 50 µm, (3, 4 and 7) = 100 µm, (5 and 6) = 150 µm.
Figures 8-13. Histological sections of testes at different maturity stages of *M. liza* from Sepetiba Bay: (8) immature testis containing spermatogonia; (9) arrangement of primary spermatocytes forming cysts (maturation stage); (10) arrangement of different testicular cells (maturation stage); (11) maturation stage after recovering stage; (12) spreading pattern of spermatozoa in a ripe/running ripe testis (spermatids are also present in small quantities); (13) recovering testis with seminiferous tubules empty and looser. (sg) Spermatogonia, (ps) primary spermatocytes, (ss) secondary spermatocytes, (st) spermatids, (sz) spermatozoa, (stw) seminiferous tubules wall. Scale bars: 25 µm.
Table II. Microscopic description of gametogenesis in *M. liza* from the Sepetiba Bay.

<table>
<thead>
<tr>
<th>Classes of LT (mm)</th>
<th>Testes</th>
<th>Ovaries</th>
</tr>
</thead>
<tbody>
<tr>
<td>300-350</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>350-400</td>
<td>3</td>
<td>7</td>
</tr>
<tr>
<td>400-450</td>
<td>31</td>
<td>35</td>
</tr>
<tr>
<td>450-500</td>
<td>46</td>
<td>64</td>
</tr>
<tr>
<td>500-550</td>
<td>6</td>
<td>15</td>
</tr>
<tr>
<td>550-600</td>
<td>2</td>
<td>13</td>
</tr>
<tr>
<td>600-650</td>
<td>0</td>
<td>12</td>
</tr>
<tr>
<td>650-700</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Total</td>
<td>89</td>
<td>154</td>
</tr>
</tbody>
</table>

DISCUSSION

*Mugil liza* testes are classified as the unrestricted spermatogonial type with cystic spermatogenesis, according to the description of Grier (1981). We have arrived at this classification because spermatogonia occur throughout the seminal lobules, and the spermatocytes consist of a group of synchronously
Reproductive biology of the mullet *Mugil liza* in a tropical Brazilian bay

The development of ovarian tissue can be divided into two phases, as in other teleosts (Wallace & Selman 1981, Forberg 1982, Howell 1983, Andrade-Talimelli et al. 1996, Solomon & Ramnarine 2007). During the first phase – the previtellogenic phase – growth is comparatively slow and there are few cytoplasmatic changes. The second phase – the vitellogenic phase – is characterized by faster growth and the deposition of large amounts of yolk in the ooplasm.

Oocytes in the previtellogenic stage (germ cells, young oocytes, or peri-nucleolus oocytes) do not contain yolk and constitute a “reserve fund” for future breeding seasons. The appearance of oil droplets in the cytoplasm (lipid vitellogenesis) is characteristic of the beginning of the vitellogenic phase and indicates that the oocytes will normally continue their development through the remaining stages within the current breeding season. The next stage, lipid and protein vitellogenesis, is characterized by the appearance of “true” yolk vesicles in the cytoplasm of oocytes. The oocytes increase considerably in size and the yolk accumulates. Vitellogenesis ceases once the oocytes reach their fully developed size, and eventually undergo maturation and ovulation after appropriate hormonal stimulation (Masui & Clarke 1979).

Histological analyses indicated that the *M. liza* exhibits synchronous group oocyte development because at least two populations of oocytes can be distinguished in the ovary at the same time during the reproductive cycle (Wallace & Selman 1981). Two clutches of oocytes were present in the ovary of specimens of *M. liza* prior to spawning: a fairly synchronous population of larger oocytes (defined as a “clutch,” i.e., post-vitellogenic oocytes) and a more heterogeneous population of smaller oocytes from which the clutch is recruited (germ cells, young oocytes, or peri-nucleolus oocytes). The former are the oocytes to be spawned during the current breeding season, while the latter are the oocytes to be spawned in future breeding seasons.

**Figures 14-15.** (14) Monthly changes in rainfall (▲), as accumulated mm, and in gonadosomatic index (\(I_g\)), hepatosomatic index (\(I_h\)) and condition factor (\(K\)), as mean ± s. e., for 89 males (■) and 154 females (○) of *M. liza* from Sepetiba Bay. (15) Monthly changes in the percent of maturity stages for female (above) and male (below) *M. liza* from Sepetiba Bay. Samples sizes are given above the bar.
In general, the ovaries of multiple spawners have both postovulatory follicles and vitellogenic oocytes simultaneously, after which the postovulatory follicles disappear gradually as the vitellogenic oocytes develop (Hunter & Goldberg 1980, Hunter & Maciejewicz 1985, Moriyama et al. 1994). Histological analyses of the ovarian tissue of *M. liza* did not show such a pattern. Instead, only vitellogenic and peri-nucleolus oocytes (ripe ovaries) or peri-nucleolus oocytes and empty follicles (after the reproductive period in recovering/resting ovaries) were observed. Therefore it can be concluded that *M. liza* from the Sepetiba Bay is a total spawner and has a similar reproductive pattern to other total spawners in *Mugil*, such as *M. platanus* (Andrade-Talmelli et al. 1996, Romagosa et al. 2000) and *M. cephalus* (McDounough et al. 2003).

Hydrated oocyte stages and spent ovaries were absent from our histological analysis samples. This may be due to the fact that our samples were collected in the inner zone of the Sepetiba Bay, a rearing and feeding ground for mullets, as described by Silva & Araújo (2000). Because mullets spawn offshore (Jacot 1920, Anderson 1957, Ditty & Silow 1996), the small proportion of mature and spent fish in the bay is likely due to the migration of adults to offshore spawning grounds. Moore (1974) also reported that during the spawning period, fully ripe fish are rarely found in coastal and embayment collections.

*Mugil liza* males are mature (*l*_{100}) at 550 mm, while females have a *l*_{100} = 570 mm. In almost all mullet species, males mature earlier than females (Salem & Mohammad 1983, Gelday 1977, Okumus & Bascan 1997), and it appears that the *M. liza* from the Sepetiba Bay is an exception. Females outnumber males in all size classes, but significant differences in the sex ratio were detected only for sizes larger than 500 mm *L*. The ratio of male to female showed an overall proportion of 1.0 male:1.73 female. It is accepted that the sex ratio is balanced (NIKOLSKY 1963) reported that shifts in the sex ratio could occur among populations of the same species and between different periods in a given population; he argued that this behavior is generally an adaptation that assures the predominance of females when environmental conditions are favorable for the production of eggs, or when the species suffers intensive fishing pressure. Since the latter is the case for *M. liza*, as for other mullets it is reasonable to hypothesize that overfishing could play a role in the predominance of *M. liza* females in larger size classes.

*Mugil liza* showed a short reproductive period ranging from May to August. The periodicity of this mullet’s reproduction may be related to environmental variability in the signals for optimal early growth and survival. Stability of the water column and suitable food in coastal lagoons, river deltas, and estuarine mangrove areas have been identified as important factors influencing the reproduction and recruitment of juvenile Mugilidae (Yañez-Arancibia 1976, Blaber & Blaber 1980, Blaber 1987, Vieira 1991). *Mugil liza* spawns during the dry season when the water column and environmental conditions are stable in the Sepetiba Bay. Early juveniles could take advantage of the stable water conditions and abundant food resources that are available in enclosed areas, such as embayments, all year round (MacGregor & Houde 1996), Silva & Araújo (2000) reported *M. liza* recruitment from May to October in the Sepetiba Bay, and these findings are in accordance with the spawning period observed in the present work.

Fecundity in *M. liza* is high, ranging from 241 x 10^4 to 365 x 10^4 compared with the co-occurring *M. curema* that shows fecundity between 82 x 10^3 and 378 x 10^3 oocytes. Other mullets, such as *M. platamus* (fecundity = 55 x 10^4 to 236 x 10^4) and *M. cephalus* (fecundity = 213 x 10^3 to 389 x 10^3), also have high fecundity (Romagosa et al. 2000, McDounough et al. 2003, Ibáñez Aguirre & Gallardo-Cabello 2004). Fecundity may vary as a result of different adaptations to environmental habitats. In the present study, high fecundity may be associated with the offshore spawning and lack of parental care that are typical of mullets. High fecundity would be a tactic to enable success in *M. liza* recruitment to the Sepetiba Bay. These results closely match the findings of Silva & Araújo (2000), who recorded large numbers of early juveniles at the inner zones of the bay.

The *I*_{p} and *K* have been used to assess fish condition and to relate this condition to reproduction. Several authors have used these parameters, coupled with *I*_{p}, to assess the reproductive period (Abascal et al. 2004, Kanak & Tachihara 2008), possibly because vitellogenesis and gametogenesis mobilize hepatic energy and body fat (Abascal et al. 2004, Kanak & Tachihara 2008). Prior to sexual maturation, marine fish generally accumulate large lipid deposits, primarily triacylglycerols, which are subsequently mobilized to support gonad development and spawning migration (Bell 1998). The major lipid storage sites are the mesenteric tissue, muscle, liver, and subdermal fat layers (Ackman 1980). In *M. liza*, the *I*_{p} was positively associated with the *I*_{p}, indicating that the liver increases in mass during the reproductive season. Increasing hepatocyte numbers and size is linked to vitellogenesis, since the provisioning of eggs with yolk takes place in the ovaries but the precursors of the yolk are synthesized in the liver (Wootton 1990). Additionally,
the lowest values for the $I_H$ were obtained after the end of the spawning season at the time when physical fatigue is greatest and fatty acid reserves are diminished. In conclusion, the $I_H$ and $I_C$ can be used together to predict the reproductive period of *M. liza* in the Sepetiba Bay.

In this study, the $K$ was not closely associated with the $I_C$. This result may suggest that reproduction does not influence fish condition, which was calculated according to the eviscerated weight of the individuals in this work. Fish in the pre-spawning period (from November through February) had a high concentration of visceral fat (not measured). This phenomenon suggests that visceral fat bodies are likely to be mobilized in late autumn for the purpose of reproduction. **Kanak** & **Tachiara** (2008) reported that decreasing visceral fat is associated with vitellogenesis in females. **Arascal et al.** (2004) concluded that fat-body lipid reserves provide an important energy source for gametogenesis in tunas. A lipidosomatic index ($I = \text{Fat Weight} \times W^{-1}$) could describe the relationship between energy depletion and vitellogenesis better than the $K$.

Our contribution provides important information on the reproductive biology of *M. liza* that will be helpful in similar studies. Further coordinated laboratory and field studies on the the frequency of spawning, fecundity, and spawning grounds of the same species are necessary for a clearer understanding of the frequency of spawning, fecundity, and spawning grounds of the same species are necessary for a clearer understanding of the reproduction of this mullet. Based on the biological findings of the present work, it is reasonable to propose that closing the fishing season for *M. liza* from May to August and setting a minimum size for capture of 350 mm L$_T$ would not only conserve stocks but also increase future harvesting in the Sepetiba Bay.

**ACKNOWLEDGEMENTS**

We wish to thank Armando Sales and Thatiana P. Ribeiro for being so helpful with the histological analyses. This research was partially funded by CAPES – Brazilian Agency for Higher Studies and Personal Graduation.

**LITERATURE CITED**


Editorial responsibility: Rosana Mazzoni.

ZOOLOGIA 27 (3): 331–340, June, 2010