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Reproductive aspects of yellow fish *Girardinichthys multiradiatus* (Meek, 1904) (Pisces: Goodeidae) in the Huapango Reservoir, State of Mexico, Mexico

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Abstract: The sexual maturity, age at first maturation and fecundity in females of the yellow fish *Girardinichthys multiradiatus* were analyzed in the Huapango reservoir located in the State of Mexico, Mexico. From July 2007 to May 2008 bimonthly samplings were carried out and, using a bait well net, 407 individuals were collected (245 females and 162 males). Overall, the sex ratio between females/males was 1.51:1 ($P<0.05$). The age of first maturation in the females was 33 mm of standard length. The spawning period occurred in July and accounted for the highest values in the gonadosomatic index. For the fertility model only fertilized eggs and embryos were considered, and those were adjusted to the potential model $F=1E\text{-}08L^{5.6144}$ ($P<0.05$). This study contributes to the understanding of the reproductive biology of the yellow fish, which takes place in the reservoirs of the State of Mexico.

Keywords: *Girardinichthys Multiradiatus,* Sex Ratio, Fecundity, Sexual Maturity

1. Introduction

Goodeidae family gathers 42 species of Mexican endemic freshwater fish, characterized among other things by presenting marked sexual dimorphism, premarital courtship and viviparity, phenomena that involve a series of morphological, anatomical and physiological adaptations, that are characteristic of the group [9]. Goodeidae family is one of the richest in endemicity and represent 32% of the endemic species of the Mesa Central of Mexico [8, 10], of these, *Girardinichthy multiradiatus* is commonly known as "the Lerma mexcalpique" or "yellow fish", and it is distributed in the top of the basins of the rivers Balsas, Lerma-Santiago and Panuco in the states of Mexico, Michoacán and Querétaro and that, according to the red List of Threatened Species [14], is listed as an endangered species.

In the State of Mexico, the environments in which this species is located are systems with little movement in its waters, with riverside or submerged vegetation and depths lesser than a meter, in addition to ponds or ditches [19]. These features coupled with urban settlements, agricultural activities, water extraction and introduction of exotic species, have put at risk the native species of this region, which has led to the disappearance of the fauna and flora in some lakes and reservoirs [31, 6]. Despite being an endemic species whose habitat is at risk due to human activities, *G. multiradiatus* has been little investigated, except for feeding, and only a few researchers have considered it threatened [4] and others think that it is not at risk [8]. In this sense, it has been mentioned that the existing information on the reproductive biology and ecology of the species is scarce, limiting the knowledge of this species in order to develop conservation and/or domestication strategies for commercial purposes [32].

Among the studies that have been made about *G. multiradiatus* we can mention behavior, feeding, and biology, but very few about its reproductive biology have been carried out; among the most remarkable works we can highlight the ones of Reference [16, 3, 17, 5, 22, 32, 21, 6, 7]; for other goodeids we can include the studies of Reference [18] and Reference [20], in addition to those reported by Reference [33], [34] and Reference [35] in their compilation of viviparous fishes.

In this regard, Reference. [33] mentioned that the study of reproduction biology constitutes an essential aspect in understanding animal species, since this aspect defines their permanence in the habitat and the balance of their populations; that is the reason why the objective of this study was to contribute to the knowledge of the reproductive aspects of *G. multiradiatus* in Huapango reservoir in the State of Mexico, and obtain information that will allow the conservation of this endemic species of Mexico.

2. Material and Methods

2.1. Study Area

Huapango is a reservoir located in the State of Mexico, in the municipality of Timilpan, between Longitude W 99° 42 '07" and Latitude N 20° 00' 24", at an altitude of 2623 masl (meters above sea level). The predominant climate is temperate sub-humid with summer rains, and it is considered one of the wettest in this category, as its total annual rainfall exceeds 800 mm of rainfall level; in addition, the prevailing winds are from north to south, and the average annual temperature is 13.8° C, with a maximum of 36° C and a minimum of -4° C with frosts from October to May. In this region, there are two very distinct climatic seasons: the dry season from November to March and the rainy season from April to October [28].

The sampling site was selected according to the criterion of Reference. [19]. On the chosen site, bimonthly samplings were carried out from July 2007 to May 2008 and in each of them an area of 13.5 m^2 on submerged vegetation was sampled, the trawling was made with a bait well net with dimensions of 45 x 25 cm in the mouth, depth of 30 cm and 2 mm mesh; the captured specimens were fixed in formaldehyde 10% [2]. All organisms were sexed, measured in its pattern length (± 0. 1 mm) and weighed on a digital balance (± 0.001 g). With the data obtained, the gonadosomatic index (GSI) and condition factor (CF) were calculated according to the criterion of Reference [24]. The sex ratio was calculated using monthly data and the degree of significance was established using the Chi-square test (X^2) with $p < 0.05$ [38].

For the elaboration of growth and fertility models, all organisms were grouped into size frequency intervals of 3.0 mm [6]. After the dissection of the incubation chamber, they were separated and counted the immature eggs (oocytes), mature eggs (ova), embryonated eggs and embryos. The fertility model was obtained with the data from embryonated eggs and embryos, which was adjusted to the potential model $F = aL^b$ [13, 27, 37]. Finally, d the logistic equation $P = 100/1 + e^{a + bL}$ was used to calculate the size of the first reproduction [1, 26].

3. Results

407 individuals were captured, 245 females (60.20%) and 162 males (39.80%) with a sex ratio of 1.5 females for each male and only in the months of July and March the sex ratio was reversed (Table 1). The length and weight averages obtained for the females during the sampling period are shown in table 2, in general the larger sizes were collected during March and July and the smaller during September. The monthly gonadosmatic index is shown in Fig. 1; the highest values correspond to July and September and the lowest November. Fig. 2 shows the monthly Condition Factor observed maximum values during July, May and November. Fig. 3 shows the percentages in the monthly variation of the stages of maturity; immature eggs (oocytes), mature eggs, fertilized (with embryo) eggs and embryos, the values observed during September and November correspond to immature fish. Fig. 4 shows the percentages by size for stages of immature eggs, mature eggs, fertilized eggs and embryos, note that embryos begin to appear in the size of 30 mm. The logistic model of Fig. 5 shows the size of first maturation which was 33 mm, which is consistent with that observed in Fig. 4. On the other hand, the model of fertility which is shown in Fig. 6 was adjusted to the potential form $F = 1E\text{-}08L^{5.6144}$

Table 1. *Total number of individuals and monthly sex ratio.*

	Females	Males	Sexual % F/M	Expected	X^2
July	39	47	0.83	43	0.37
September	46	10	4.60	28	11.57
November	26	15	1.73	20.5	1.48
January	53	31	1.71	42	2.88
March	28	31	0.90	29.5	0.08
May	53	28	1.89	40.5	3.86
Total	245	162	1.51	203.5	8.46

Table 2. *Monthly averages in length and weight for females*

	Length (mm)			Weight (g)				
	minimum	mean	DS	maximum	minimum	mean	DS	maximum
July	34.00	41.35	4.12	49.00	1.45	2.21	0.66	3.43
September	8.70	15.85	3.96	23.10	0.01	0.11	0.11	0.28
November	16.50	22.75	3.97	34.80	0.62	0.22	0.09	0.95
January	15.00	20.34	2.54	28.40	0.08	0.17	0.07	0.43
March	18.00	22.84	2.57	27.40	0.10	0.24	0.09	0.40
May	20.20	25.16	4.17	45.50	0.19	0.44	0.34	2.55

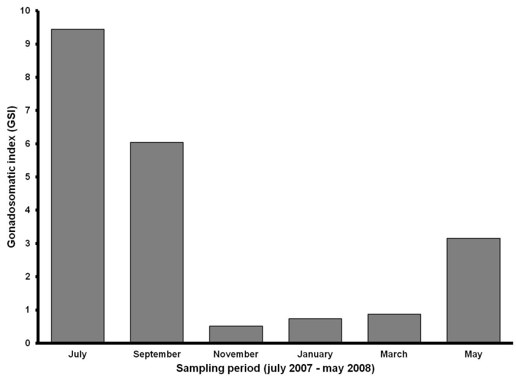

Figure 1. *Values of gonadosomatic index (GSI).*

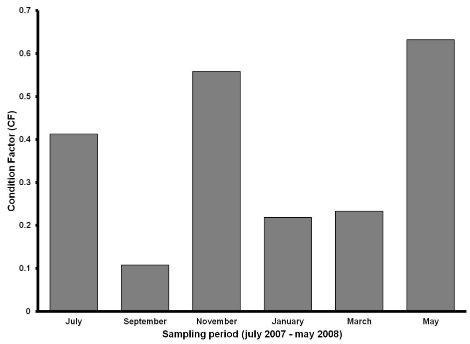

Figure 2. *Monthly values of Condition Factor (CF).*

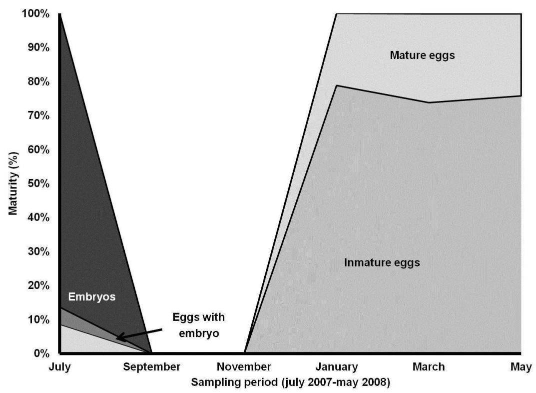

Figure 3. *Percentages of monthly maturation of immature eggs stadiums, mature eggs, fertilized (with embryo) eggs and embryos.*

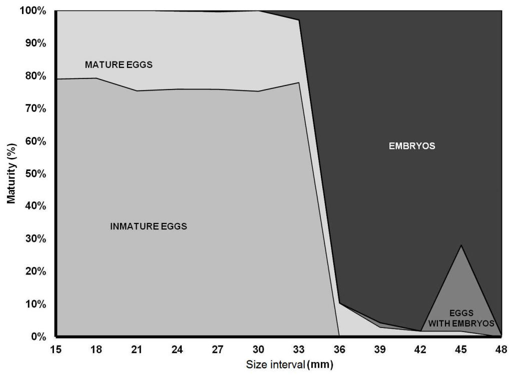

Figure 4. *Percentages of maturity size obtained during the sampling period.*

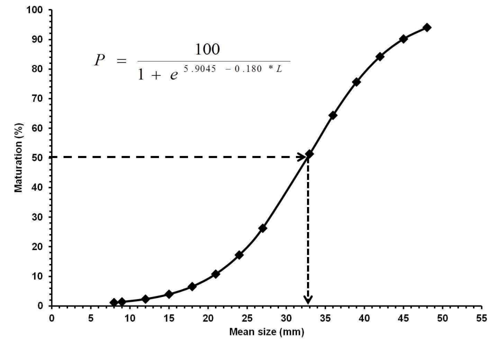

Figure 5. *Logistic model for obtaining the size of G. multiradiatus at first reproduction.*

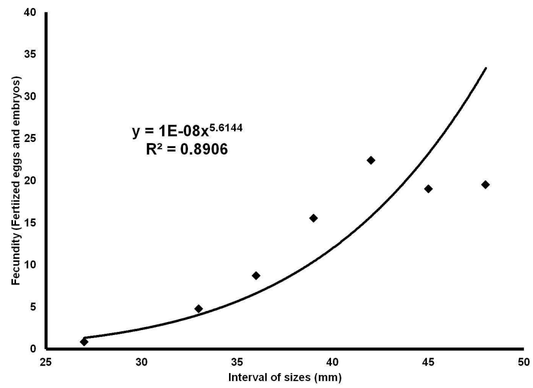

Figure 6. *Fertility model of G. multiradiatus obtained from the data of eggs with embryo and embryos.*

4. Discussion

Overall, the sex ratio favored to females; however this proportion changes monthly as shown in Table 1, which shows July and March with the sex ratio favoring males (0.83 and 0.90 respectively). Reference [6, 7] reported a similar sex ratio for this species in reservoirs of San Martín in the state of Querétaro and in Villa Victoria, State of Mexico (1.7:1 and 2.27:1 respectively). The sex ratio favoring females is also reported for other goodeids; of 1.5:1 and 4:1 for *Hubbsina turneri* De Buen, 1941 [20], but for *Zoogoneticus quitzeoensis* Bean, 1898, this ratio was 1:1 [23]. Another report, in which the sex ratio in general favors males, was recorded at the reservoir of San Miguel Arco, State of Mexico with sex ratio males/females of 1.39:1 [21]. For other ovoviviparous species Reference [12] reported a sex ratio of 1.7:1, in favor of females in *Heterandria bimaculata* Heckel, 1848. In this regard, Reference [30] mentioned that in Poeciliids (viviparous and ovoviviparous species) the sex ratio in general favors females in wild populations, which allows them to ensure the reproduction. But he also mentions that the changes related to the sex ratio may be due, among other things, to the size of the males and their feasibility in the catch, to the segregation in the habitat by sex and to the predation toward males; the latter was also reported by Reference [15] which might happen to this species and explain the different values of the reported sex ratio for this species. In this sex ratio also influences the use of different trapping methods used in various systems, where seines or bait well nets are used.

Furthermore, the corporal indices that have been introduced searching for a simple and objective criterion to quantify the gonadal development are useful for identifying the time of spawning, especially in species which have a marked seasonality [26] which apparently is the case with this species. In this case, the highest values of the GSI, also called coefficient of maturity (9.43 in July, 3.15 in May), correspond to the sizes of larger females, captured in these months (41.35 mm in July, 25.16 mm in May), as well as also with values higher in the CF (0.41 in July, 0.63 in May), months in most big and pregnant females were captured (Figs. 2 and 3). Similar values are reported for *Ameca splendes* [22] and *H. turneri* [20]. Reference [26] also mentions that seasonality may be more marked in systems water, less stable, as it occurs in the reservoirs in the State of Mexico, which are subject to a variation in their volumes due to the extraction of water for irrigation.

In this paper, the results indicate a breeding season for the yellow fish from May to September (Fig. 3), coinciding with data reported by Reference [21] in the Reservoir of San Miguel Arco in the State of Mexico and Reference [6, 7], in San Martin, Querétaro and Villa Victoria, State of Mexico. These are also the months where, in addition, the highest values of GSI are observed (Fig. 1). For other goodeids species, Reference [18] and Reference [22] report a reproductive season similar to the *G. multiradiatus*. However, these data differ from those reported by Reference [19] who reported a period of reproduction from December to May, while Reference [17] mentioned that the reproductive cycle of this species has a period of continuous reproduction. However, in none of these cases it is mentioned whether the data correspond to annual samplings as the ones of the present paper. In this and in the above mentioned reservoirs, the reproduction is interrupted from November to February, month in which the lowest temperatures are registered in the area, ranging from 9-12° C, corresponding to the dry season [29]. This information is consistent with what Reference [9] reported for *Girardinichthys viviparous*; they indicate that during these months an interruption in the reproduction of this species can be observed due to environmental effects. In contrast, Reference [12] indicate that the maximum period of viviparous reproduction for some fish is related to the increase in the water level and temperature, as it occurs in these bodies of water and in this region in particular, which temperature and water level begin to increase from May to October, representing the rainy season in the area [29].

On the other hand, the average size observed along the sampling was 24.95 mm, similar to the one observed in the reservoir of San Martín (20.4 mm). This is because during the dry season is more evident the absence of adult specimen, due to the low water levels of these systems, especially in San Martin which is emptied almost in 90% [6].

The maturity percentages show that the gonads of the organisms with size range of 15-30 mm (Fig. 4) begin to mature or have already matured and, moreover, the apparition of fertilized eggs and embryos in organisms with size of 33 mm (10 embryos in average) suggests that this is the size of maturity onset, and this is evidenced in the model obtained from the maturity data. Reference [6, 7] found that the size of *G. multiradiatus* at first reproduction in the reservoir of San Martin, Querétaro was of 30 mm with 18 embryos on average and in Villa Victoria, State of Mexico, was of 32 mm also with 18 embryos on average. The maximum number of embryos observed in this study was 68, in sizes of 47 mm, while Reference [21] reported 87 embryos in San Miguel Arco. For other goodeids, Reference [23] reported for *Z. quitzeoensis* a maturation size of 30 mm, but an average of 10 embryos; this is the lowest average for goodeids. Reference [18] reported an average of 20 embryos for *Allophorus robustus* and *Goodea luitpoldii* and 40 embryos in average for *Neophorus diazi*. Furthermore, Reference [34] reported an average of 30 embryos in *Ilyodon whitei* and 55 in *Goodea atripinnis*. In this regard, Reference [26] mentions that the age and size of first maturity can vary within the same species, and that this variation may be spatial or temporal; in addition, he refers to the fact that populations of the same species have different values in different areas of their distribution range, as it has been mentioned for the different systems in which *G. multiradiatus* has been studied.

The fertility model (Fig. 6) showed similar information to what has been reported for *G. multiradiatus* in the reservoir of San Martin, Querétaro [6], and shows a direct relationship between the number of embryos and the size of the organisms. In this sense, the fertility model as it applies to the oviparous species was not applied to *G. multiradiatus,* given the viviparous characteristics of this species. In this type of fishes is common to observe a decrease in the number of eggs as sexual maturity advances to give way to the number of embryos to be born, due to which fecundity (fertility) in these organisms should be referred to the complex consisting of embryonated eggs and embryos at different stages in development found at the time of capture and preservation. According to this, it is better to refer to the concept of fertility [27, 12, 36], for viviparous and ovoviviparous species, where the effective number of embryos to be born and their relation to the size are shown. However, more studies on fertility in viviparous and ovoviviparous species need to be considered in order to establish differences or similarities in the reproductive behavior of these species and to establish the proposed model of fertility.

The results in this study do not differ from those reported for the reservoirs of San Martin and Villa Victoria, both in CF and GSI (Figs. 1 and 2), which reflect the physiological conditions of the yellow fish and showed a direct relationship with the female size and its maturity; this variation indicates that the reproductive period of the yellow fish occurs from

May to September (Fig. 3), because after the reproductive period, there is an increment of immature organisms and organisms in maturing process with sizes that are smaller than the size of first maturity (33 mm) (Figs. 4 and 5). Likewise, values of fecundity and size at first sexual maturity, as mentioned above, differ slightly from the different systems studied.

GSI values reflect the development of the gonads and could well be used as an indicator of gonadal maturation, just as the CF that is related to GSI could be considered as a complement to gonadal maturity stages in this species. These data suggest that both the GSI and CF (Figs. 1 and 2) can be used to determine the degree of maturity and identify the time of egg-laying of the yellow fish, as mentioned by Reference [26] for species of seasonal egg-laying and, in this case, a seasonal breeding period can be clearly observed (Fig. 3). Studies on these parameters (CF and GSI) were carried out in *Ameca splendes* [22] and *H. turneri* [20]. They also show a relationship with maturity stages, similar to those reported in this study for *G. multiradiatus.*

The above mentioned is indicative of the particularity and dynamical environments in which some freshwater species develop. As in this case, it is likely that variations in the water level due to the use of water for irrigation during the dry months, coupled, among other factors, with climatic seasons and productivity in systems, may influence the reproductive aspects of the yellow fish, not only from a physiological point of view, but also from a behavioral standpoint. This applies not only to this species but to most goodeids subject to habitat modification and whose geographic range is restricted [8].

The conservation of goodeids is an important work that should be done, but it seems difficult due to the scarcity of studies on reproduction of the species and to the irrational use of natural resources that damage their habitat [11]. In this sense, some authors mention that aquaculture can be considered as a tool for the conservation of endemic and endangered native species, but based on the knowledge of their biology and establishing techniques that allow their cultivation [25], studies of this nature are necessary to protect endangered species.

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