



Life history traits of *Xiphophorus kallmani* Meyer & Schartl, 2003 (Pisces: Poeciliidae)

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Abstract. *Xiphophorus kallmani* Meyer & Schartl, 2003, is a freshwater fish endemic to Lake Catemaco, Veracruz, Mexico, whose populations are currently threatened by habitat degradation, pollution, and collection for the aquarium trade. Given its restricted distribution and vulnerable status, information on its life-history traits is essential for conservation and management efforts. This study describes key reproductive and developmental traits of *X. kallmani* under laboratory conditions. Broodstock groups were maintained in recirculating aquaculture systems, and offspring were monitored from birth until sexual maturity. Brood size, sex ratio, age at maturity, body weight (BW), total length (TL), fecundity, and fertility were recorded and analyzed using Pearson correlation analysis. The mean brood size was 79.5 ± 4.7 offspring per litter, with a strongly female-biased sex ratio. Males reached sexual maturity earlier than females, a pattern consistent with other poeciliid species. Significant positive relationships were observed between female fecundity and fertility, between age and BW at maturity in males, and between male TL at maturity and female fertility. These results provide the first baseline data on the life-history traits of *X. kallmani* and offer valuable information for the development of ex situ breeding, management, and conservation programs for this threatened endemic species.

Key Words: reproductive biology, life -history traits, Poeciliidae, endemic fish, conservation aquaculture.

Introduction. Mexico is a country that stands out for its large number of poeciliid species, being the fish family with the largest number of species registered in its territory (105 species) and being the country with the largest number of species of the genus *Xiphophorus* registered worldwide with 89.28% of the total species registered, however despite its great biodiversity more than half of its species are threatened and within some category of protection (Hernández-López et al 2022).

In this context, *Xiphophorus kallmani* Meyer & Schartl, 2003 is an endemic fish to Catemaco, Veracruz. Its population is concentrated in Lake Catemaco, which in recent years has suffered transformation of its surrounding habitats due to excessive logging, the presence of pesticides, and water pollution. Adding to this is the direct threat to *X. kallmani*, its extraction and capture for aquarium purposes (Lorán et al 2013). For these reasons, its populations have been affected, and it is considered a threatened and vulnerable species according to the classification of North American freshwater fishes (Jelks 2008). It is especially sensitive to extinction due to its restricted geographic range (Johnson & Hubbs 1989). Therefore, there is a need to understand aspects of its life history, which are important for breeding and conservation purposes in ex situ aquaculture programs, as has been done with other threatened native poeciliids (Hernández-López & Luna-Vivaldo 2025).

This study aimed to describe key reproductive and developmental life-history traits of *X. kallmani* under controlled laboratory conditions, providing baseline data for conservation and ex situ breeding programs.

Material and Method

Broodstock. The study was carried out in the Faculty of Veterinary Medicine and Animal Husbandry of the Veracruz University in Veracruz, México, from October 2021 to October 2022.

45 juvenile fish were collected (12 males and 33 females) from Lake Catemaco. Females and males were kept separate for six months in different tanks until they reached maturity. After this time from these individuals, ten breeding groups were established at a 1:3 (male: female) ratio, following the guidelines of Peña & Ma (2001). The males exhibited a mean body weight (BW) and total length (TL) of 3.62 ± 0.11 g and 7.20 ± 0.34 cm, respectively, while females showed mean values of 4.55 ± 0.85 g and 7.61 ± 0.41 cm. Each breeding group was housed in a 40-L tank connected to a recirculating aquaculture system equipped with mechanical and biological filtration.

Each tank was fitted with a mesh cover and artificial plants to provide shelter and prevent parental cannibalism, as well as additional environmental enrichment elements (Webster 2001; Llanos & Scotto 2014; Cyprinodontiformes et al 2024). The broodstock were maintained in the recirculating system for two months, during which fry were obtained from eight of the ten breeding groups. The fry were then separated by brood and transferred to a second recirculating system, where they were kept until reaching sexual maturity.

Culture conditions. The study was carried out with a 12:12-hr light: dark photoperiod, and water quality parameters were maintained within ranges suitable for fish farming and consistent with values reported in previous studies involving poeciliid fish from Lake Catemaco (Ramírez 2014). Water parameters were measured twice weekly with a Nutrafin® multiparameter water sensor and a colorimetric test (Table 1). Water was exchanged weekly, and evaporated water was replaced daily to prevent abrupt changes in water parameters.

Water quality parameters measured during the study

Table 1

Parameter	Range value
pH	7-7.5
NO ₃ (mg L ⁻¹)	0
Ca (mg L ⁻¹)	20-25
NH ₃ (mg L ⁻¹)	0
NO ₂ (mg L ⁻¹)	0
GH (mg L ⁻¹)	95±5
KH (mg L ⁻¹)	70-85
Fe (mg L ⁻¹)	0
PO ₄ (mg L ⁻¹)	2-2.5
Temperature (°C)	28-30
Oxygen (mg L ⁻¹)	5.8-6

Note: GH - general hardness; KH - carbonate hardness.

Feeding. Newborn fish were fed exclusively with the nematode *Turbatrix aceti* at a concentration of 650 organisms mL⁻¹ (15 mL per tank) five times daily, following Hernández-López and Luna-Vivaldo (2021). From four days post-birth until the onset of sexual maturation, fish were fed four times daily with specialized fry powder (Ultrafresh®, 42% protein) *ad libitum* and once daily with *T. aceti* at the same concentration used for the newborns. Subsequently, from the onset of sexual maturation until the end of the study (when sexual maturity was reached), fish were fed three times daily: twice with specialized feed for guppies and livebearers (Ultrafresh®, 44% crude protein) and once with Artemia flakes (Acuabiomar®, 46% crude protein) *ad libitum*.

Litter size. To determine litter size, the average number of offspring per litter from the breeding lots was considered.

Total length, weight, and age of sexual maturity in males. To determine the age, TL, and weight at which sexual maturity was reached in males (Figure 1), biometric measurements were taken every two weeks using an analytical balance (0.01 g precision) and a digital caliper (0.1 mm precision). Measurements were taken until the complete transformation of the gonopodium was observed (Vega-Trejo et al 2019).



Figure 1. *Xiphophorus kallmani* males upon reaching sexual maturity.

To reduce stress in the fish and facilitate handling during measurement and weighing, the organisms were anesthetized (Ross & Ross 2009) with 125 (mg L⁻¹) of eugenol, following the protocol for the anesthesia of *X. helleri* (Llanos & Scotto 2010).

Total length, age of sexual maturity, and fecundity of first reproduction in females. To determine the TL and weight at which females reach sexual maturity, they were measured and weighed following the same procedure as for males, until a bulge and/or gravid spot was observed in the abdomen, indicating the presence of eggs (Figure 2). The females were then dissected (after euthanasia by overdose of the previously used anesthetic) and, with the aid of a stereomicroscope, the presence of mature ova (stage III) was confirmed. This is indicative of sexual maturity, according to Haynes (1995). To determine fecundity, the total number of ova and oocytes in stages I and II at the time the females were analyzed was considered, according to Schoenherr (1977).



Figure 2. Female of *Xiphophorus kallmani*, shown encircled in red, the gravid spot.

Determination of first reproductive fertility. Fertility was defined as the total number of embryos contained in the females at the time of dissection (Schoenherr 1977). After dissection, the organisms and eggs were preserved in 95% alcohol.

Statistical analysis. To identify potential relationships among BW, TL, litter size, fecundity, fertility, and age at sexual maturation, Pearson's correlation analysis was performed using the Statistical Analysis System[®] (SAS) On Demand for Academics Communities program statistical software (PROC CORR). Before conducting the correlation analyses, the assumptions underlying Pearson's correlation were evaluated. Data normality for each variable was assessed using the Shapiro–Wilk test, and linearity between variable pairs was examined through visual inspection of scatterplots.

All variables met the assumptions of normality and linearity, justifying the use of Pearson's correlation coefficient. The analysis provided estimates of the Pearson correlation coefficient (r), associated p-values to assess statistical significance, and basic descriptive statistics. All statistical analyses were conducted using SAS, with the significance level set at $p < 0.05$, such that results were considered statistically significant when the probability of error was less than 5%.

Results and Discussion. Brood size. One litter was obtained from each breeding group, resulting in a total of 795 offspring, of which 135 were males and 660 were females., with a ratio of 1:4.88 (males:female), which is similar to the 1:4.43 (males:females) ratio reported for *X. helleri* (Milton & Arthington 1983), The mean brood size was 79.5 ± 4.7 offspring per litter. These values are consistent with previous reports for *Belonesox belizanus*, a species in which litters of similar size have been recorded with 77.7 ± 5.04 offspring (Turner & Snelson 1984), and an average brood size of 88.1 ± 12.3 reported in *Limia vittata* (Cohen et al 2015). Comparable brood size has also been documented for *X. helleri*, with an average brood size of 84.86 ± 10.46 offspring (Llanos & Scotto 2014). Summary statistics for the evaluated life history traits are presented in Table 2.

Table 2

Life history traits of *Xiphophorus kallmani* obtained during this study

Life history trait	<i>Xiphophorus kallmani</i>	
	Male	Female
Age of sexual maturation (weeks)	22.27±3.5	24.30±2.24
Total length at maturation (mm)	51.26±4.23	50.66±7.03
Weight at maturation (g)	3.24±0.38	3.72±0.53
Fecundity (total ovas)	-	57.64±12.05
Fertility ar first reproduction (number of embryos)	-	5.46±4.33

Note: n = 135 (male); n = 666 (female).

Males of *X. kallmani* matured earlier than females, which is consistent with patterns reported for other poeciliids, in which females typically reach sexual maturity later than males, such as in *Poecilia reticulata* and *Gambusia marshi* (Meffe 1985; Mahmud-Al-Hasan et al 2025). In addition, the age at maturity observed in male *X. kallmani* was similar to that reported for male *Xiphophorus nigrensis* (mean of 22.6 weeks) (Morris & Ryan 1990). Positive correlations were found between fecundity and fertility ($p < 0.05$) in females, while in males a correlation was found between age of sexual maturation and weight at sexual maduration ($p < 0.01$), This is consistent with previous studies showing that males that stop growing until they mature, therefore those that take longer to mature will reach greater weight and length (Borowsky 1973; Reznick et al 2021).

A positive correlation was observed between male TL at sexual maturity and female fertility, measured as the number of embryos ($p < 0.05$, $r = 0.71$). This finding indicates that larger males are associated with higher female reproductive output. This relationship may be explained by size-dependent differences in sperm production, as larger males are likely to produce and transfer greater quantities of sperm, thereby increasing the likelihood of fertilizing a higher number of ova. Similar patterns have been reported in *Gambusia holbrooki*, a poeciliid species in which larger males exhibit increased sperm production (Vega-Trejo et al 2019).

Conclusions. This study presents the first detailed characterization of key life-history traits of *Xiphophorus kallmani* under laboratory conditions. Fundamental reproductive and developmental parameters were documented, including brood size, sex ratio, fecundity, fertility, and sex-specific patterns in age, BW, and TL at sexual maturity. Overall, these traits are comparable to those reported for other poeciliid species, although *X. kallmani* shows a strongly female-biased sex ratio and earlier maturation in males.

Significant relationships between growth-related traits and reproductive output highlight the role of body size and age at maturity in shaping reproductive performance. In particular, the positive association between male TL at maturity and female fertility suggests a potential influence of male size on reproductive success.

These results provide essential baseline information for a threatened, narrowly distributed endemic species and constitute a valuable foundation for future ex situ breeding, management, and conservation programs.

Conflict of interest. The authors declare that there is no conflict of interest.

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