Received: 23 May 2018 | Accepted: 15 September 2018

DOI: 10.1111/eff.12451

ORIGINAL ARTICLE

WILEY Ecology of FRESHWATER FISH

Comparing three methods to estimate the average size at first maturity: A case study on a Curimatid exhibiting polyphasic growth

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Funding information
Conselho Nacional de Desenvolvimento Científico e Tecnológico, Grant/Award Number: 305017/2016-0; Coordenação de Aperfeiçoamento de Pessoal de Nível Superior; Killam Predoctoral Scholarships, Grant/Award Number: 88881.119097/2016-1; Programa de Pesquisa em Biodiversidade; Vanier Canada graduate scholarships

Abstract

The average size at first |maturity (L_{50}) is among the most important parameters for fisheries management and conservation. This paper aims to compare three different methods for its estimation. Considering a classical approach, a logistic model was used (a) by determining the gonadal stage macroscopically; and (b) by using the GSI as proxy of sexual maturity; and finally; (c) by using the length–weight relationship (LWR) in a theoretical approach. The proposed methods were applied using data of a detritivorous fish, *Cyphocharax abramoides*, monthly sampled using gill nets. Captured individuals were measured, weighed, sexed and the gonadal stage was classified macroscopically and weighed. Estimated L_{50} values using the macroscopic identification, GSI approach and LWR were not different from each other considering confidence intervals. Between the three different techniques, we concluded that the analysis of the LWR in fishes with polyphasic growth presented promising results as it only requires length and weight data to be performed and estimate a L_{50} within the range of both classical logistic models analysed.

KEYWORDS

estimated parameters, fish reproduction, Gonadosomatic Index, length–weight relationship, polyphasic growth, size at first maturity

1 | INTRODUCTION

Studies on the reproductive biology of fish species provide important information for both basic fish biology and applied fisheries management and conservation (Barbieri, 1995; Marques, Braun, & Fontoura, 2007). Despite the growing need to establish management measures, many of the population and reproductive parameters that would provide useful information for adequate management decisions are still scarce (Favaro, Lopes, & Spach, 2003; Rosa & Lima, 2008). This occurs mainly because: (a) most available data for many fish species come from fishery-dependent sources, which usually presents a better description of adult fishes; (b) a general lack of standardised sampling for fish sampling gears and effort; and (c) no monthly samples, which precludes quantifying the reproductive period for many fish species and can bias reproductive parameter analyses (Booth, 2000; Gavaris, 1980; Safran, 1992; Ticheler, Kolding, & Chanda, 1998).

The average size at first maturity (L_{50}) describes the individual body size at which 50% of a population is mature and is one of the most important parameters for fisheries management. L_{50} assesses the maturity and reproductive schedule of a population as a trade-off to plastic (i.e., variable) lifetime growth patterns (Lorenzen,

2016) and allows for estimates of the maximum length of a species, minimum catch size, and therefore appropriate size restrictions for fisheries-dependent gears (e.g., mesh size of fishing nets and the appropriate hooks size for fishing lines) (Schill, LaBar, Mamer, & Meyer, 2010; Shephard & Jackson, 2005; Stark, 2012).

Classical methods for estimating L_{50} of fishes range from the *probit* method to multivariate and logistic models that use the length or age according to a proportion of mature fishes identified macroscopically or using Gonadosomatic index (GSI) as proxy of sexual maturity (Chen & Paloheimo, 1994; Fontoura, Braun, & Milani, 2009; Richards, Schnute, & Hand, 1990). Both these approaches (macroscopic and GSI) are classically accepted in fisheries management as providing accurate and unbiased estimates of L_{50} (ICES, 2008); however, both of these methods are lethal for individuals sampled for maturity (Crim & Glebe, 1990), and cost time in terms of intensity of sampling and technical laboratory work.

A promising nonlethal method to estimate L_{50} auxiliary to either the macroscopic or the GSI sampling approach is to consider the length–weight relationship (LWR; $W = a.L^{b}$) described by Huxley (1924). LWR considers that the relationship between weight and length is constant throughout life, meaning that body dimensions increase relative to each other according to an allometric coefficient constant (b) in a monophasic growth. However, fish often go through different stages of growth, as at the beginning of its development when abrupt physiological changes occurs (Vergara, Sigurdsson, & Saborido-Rey, 2013). A fundamental change in somatic growth patterns of most fish species is the change in energy allocation when individuals of a population reach sexual maturity, since there is an investment in gonadal development (Quince, Abrams, Shuter, & Lester, 2008a,b; Shuter et al., 2005; Wilson, Honsey, Moe, & Venturelli, 2018). Hence, a change point in allometry over the lifetime LWR of individuals of a population can correspond to the size at which individuals mature, reflecting a polyphasic growth.

Bervian, Fontoura, and Haimovici (2006) reviewed the original Huxley (1924) equation using three different approaches to develop a statistical model for accurate estimates of relative growth. As a result, the best fit was obtained by considering a polyphasic model where a growth pattern can be divided into at least two different phases, suggesting that a change in the growth parameters was associated with sexual maturity. Considering this polyphasic growth model, Fontoura, Jesus, Larre, and Porto (2010) tested the hypothesis that from the LWR analysis of a fish with polyphasic growth it is possible to infer the size at maturity, based on the premise that the change in energy invested into reproduction is reflected in a change in growth parameters.

Considering the quantity and the quality of available data, the objectives of this study were to compare three different methods of estimating $L_{50,}$ aiming to identify which one is more accurate. We first considered the classical approaches: a logistic model using the total length according to a proportion of mature fishes that were classified in two different ways, (a) by determining the gonadal stage macroscopically; and (b) by using the GSI as proxy of sexual maturity. In addition, assuming that changes in allometric growth patterns can reflect the reproduction influence on the amount of energy directed for body growth; (c) we aim to test the hypothesis proposed by Fontoura et al. (2010) in order to ascertain whether weight and length data provide adequate information to estimate the average size of first maturity of fish with polyphasic growth. The proposed methods were applied using data of *Cyphocharax abramoides* (Kner, 1858), a detritivorous Amazonian Curimatidae chosen for its abundance (Ayres-Santos, Freitas, & Montag, 2018), monthly reproduction, and well identifiable gender and maturation status.

2 | MATERIALS AND METHODS

2.1 | Specimens data

Monthly samples of *C. abramoides* were taken from May 2012 to April 2013 in the lower Anapu River in Caxiuanã National Forest (1° 45′ 27.5″ S, 51° 27′ 33.2″ W), located in the Xingu-Tocantins interfluvium, Pará state, eastern Brazilian Amazon, using gill nets (knot-to-knot meshes of 15, 20, 25, 30, 35 and 40 mm and total length of approximately 200 m) submersed between 4 and 10 p.m. Collected fishes were submitted to a lethal dose of anaesthesia (Eugenol solution) following the American Veterinary Medical Association (Underwood, Anthony, Gwaltney-Brant, Poison, & Meyer, 2013).

Captured individuals were measured to obtain total length (TL) to the nearest 0.1 cm and weighted to obtain the total weight (TW) to the nearest 0.1 g. One ventral longitudinal incision was made to observe, remove and weight the gonads. The individuals were sexed and the gonadal development was classified macroscopically according to Núñez and Duponchelle (2009) in immature (A); at maturity (B); mature (C) and spawned or spermiated (D). Stage A individuals were classified as juveniles, and stages B, C and D individuals were classified as adults.

Similar to others species from the genus *Cyphocharax*, *C. abramoides* presents multiple spawning events throughout the year, with one peak related to pluviometric period (Hashiguti, Rocha, & Montag, 2017). According to Hashiguti et al. (2017), the gonads of both sexes are macroscopically different regarding colour pattern, size, consistency and vascularisation for each gonadal development stage and the macroscopic identification can be used to classify gonadal stages.

Voucher specimens were deposited in the ichthyology collection of the Museu Paraense Emílio Goeldi (MPEG; Belém, Pará, Brazil) under MPEG 30477 and MPEG 30478 codes.

2.2 | Statistical analysis

The L_{50} was determined using two different methods, both based on the number of adult's individuals per length class, according to the formula:

$$P = \frac{P_{max}}{1 + e^{-\ln(19)\frac{TL - L_{50}}{\delta}}} \quad (1)$$

where *P* is the proportion of adults individuals, *Pmax* is the asymptote (presumed 1.0 in a maturity model), TL is the total length (cm), L_{50} is the average length (cm) when 50% of the population is sexually mature, and δ is the average increment (cm) after L_{50} to reach 95% proportion mature (as constrained by the natural logarithm of the odds ratio 19/1). In the second method, the reproductive maturity was established using the average values of the Gonadosomatic Index (GSI) for males and females (Santos, 1978) according to the formula GSI = (*Gonad Weight/Total Weight*)*100, with 5% cut proposed by Marques et al. (2007) and Fontoura et al. (2009). The 5% cut considers as adult an individual with at least 5% of the maximum gonad weight registered.

The proportion of mature fish per size class (rounded to the nearest half centimetre; ranging from 8–22.5 cm) were used as the data in Equation 1 and we estimated the parameters L_{50} and δ by minimising the negative log likelihood using a binomial probability density function, where the number of trials are the number of fishes in each centimetre group, and the number of successes are the number of mature fishes in each centimetre group. Model fitting was done using the function *mle* in R version 3.2.3 (R Core Team, 2015) with starting values of 15 and 10 for L_{50} and δ respectively. Asymptotically normal standard errors for the L_{50} parameter were derived by taking the square roots of the diagonal elements of the inverse of the Hessian matrix from the maximum likelihood solution, and 95% confidence intervals for L_{50} were approximated by taking the maximum likelihood estimate for L_{50} ± twice the standard error of the parameter.

The potential for polyphasic allometric growth in *C. abramoides* was evaluated from the length–weight relationship (LWR) in Huxley (1924). We used the formula:

$$\mathrm{TW} = a\mathrm{TL}^{b} \tag{2}$$

where TW is the total weight (g), *a* is the coefficient of proportionality, TL is the total length (cm), and *b* is the allometric shape coefficient. After the construction of the model, the proportional residuals ((observed weight – expected weight)/observed weight) were plotted as a function of TL to identify possible trends in the data looking for a potential shift in allometric growth associated with maturity. The proportional residuals were used in place of regular residuals (observed weight – expected weight) in order to minimise heteroscedasticity present in the data. Preliminary estimates of the parameters *a* (coefficients of proportionality) and *b* (allometric shape parameter) from Equation 2 were estimated by minimising the sums of squares in the proportional residuals using the function *mle* fitted to observed length and weight data. Analyses of the residual graphs from the fitted Equation 2 showed that the proportional residuals were not randomly distributed (around 0 on the *x*-axis) along the *y*-axis. The break in the residual pattern indicated a point of change in the growth pattern, meaning that the LWR was not suitable for a regular potential equation, suggesting polyphasic growth. To confirm the existence of residuals trends along the subjects' lengths, we fitted a polynomial smoothing spline and identified where the initial change point occurred for biased residuals. The growth in different phases represents a change point in allometric growth for *C. abramoides* individuals associated with the investment into reproduction. According to Bervian et al. (2006), such phases can be described by different power equations that can be switched on and off at a change point defined as a stanza changing length.

Unlike Bervian et al. (2006), which defined the stanza changing length (SCL) with a logistic equation with two shape parameters that created the switch function between the juvenile and adult phases, we estimated the SCL parameter directly as a change point between two different length–weight relationships associated with the juvenile phase and adult phase. Hence, the following:

$$TW = \left\{ \begin{array}{l} a_1 TL^{b_1}; TL < SCL \\ a_2 TL^{b_2}; TL \geq SCL \end{array} \right\} \tag{3}$$

here a_1 and b_1 represent the coefficients of proportionality and allometric scalar for the juvenile phase, respectively, a_2 and b_2 represent the coefficients of proportionality and allometric scalar for the adult phase, respectively, and SCL is the size that represents the average length (in cm) where the second growth phase occurs. The allometric shape parameter *b* can indicate if a species has isometric (*b* = 3, fish dimensions increase at the same rate) or allometric growth (*b* ≠ 3), which can be divided in positive (*b* > 3, higher weight increase) and negative allometry (*b* < 3, higher length increase) (Froese, Tsikliras, & Stergiou, 2011; Morey et al., 2003). Suitable starting values for SCL were determined by taking the logarithm of Equation 3 and fitting a continuous piecewiselinear regression using the *segmented* package in R (Muggeo, 2008). Using the initial starting values provided by the piecewise linear model, we then fitted length and weight data to Equation 3 and estimated the parameters a_1, b_1, a_2, b_2 and *SCL* (all five parameters were constrained positive) by minimising the sums of squares of the proportional residuals using the function *mle*. Similar to above, asymptotically normal standard errors for the *SCL* parameter were derived by taking the square roots of the diagonal elements of the inverse of the Hessian matrix from the minimised sums of squares solution, and 95% confidence intervals for *SCL* were approximated by taking the minimised sum of square residuals estimate for *SCL* ± twice the standard error of the parameter. We then verify whether the stanza changing length is effective in estimating the first mature size by contrasting the mean and 95% CI for the L_{50} (macroscopic and GSI based) to the mean and 95% CI for the *SCL* from the LWR approach.

3 | RESULTS

A total of 872 *C. abramoides* specimens were captured, with 459 females and 413 males identified macroscopically and total length ranging from 9 to 22.5 cm. Females presented a mean length of 17.4 cm (± SD 3.8) and average weight of 84 g (± SD 30.5), while males showed mean length of 16.4 cm (± SD 1.9) and weight of 63 g (± SD 21.3). Mature and immature individuals were captured every month, and a high number of mature individuals were sampled (females: 80.4% and males: 44.7%).

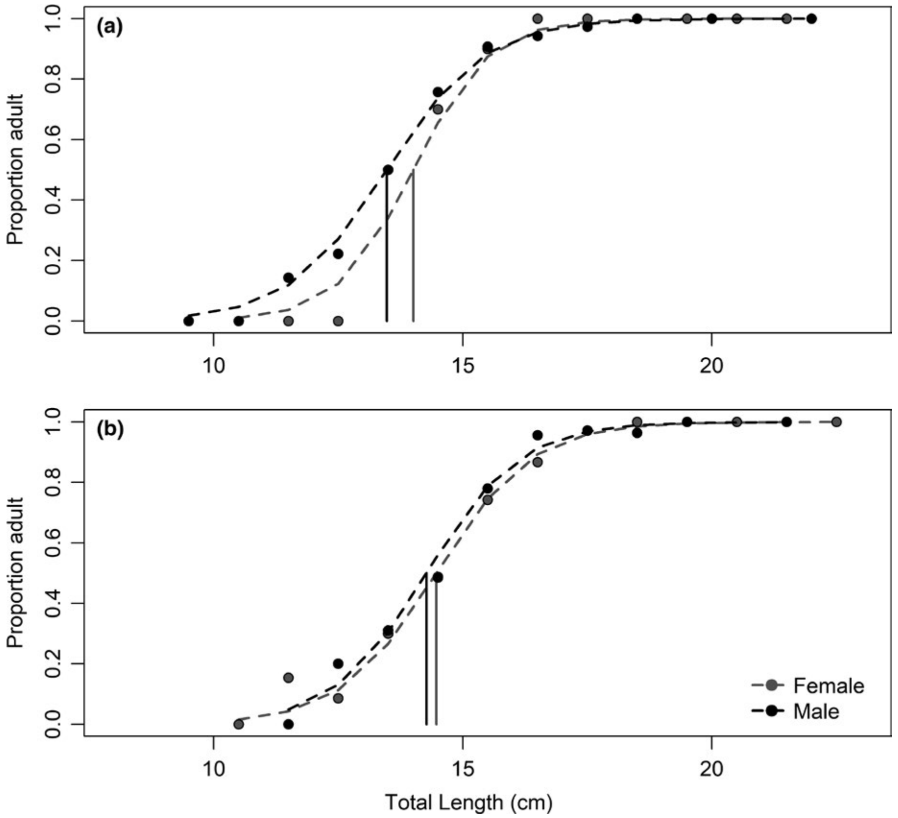

FIGURE 1 Proportion of sexually mature male and female *Cyphocharax abramoides* by body size (total length) using a macroscopic identification (a) and GSI approach (b)

TABLE 1 Comparison of three different methods for estimating the size at first maturity (total length in cm) for male and female *Cyphocharax abramoides* with associated sample size (*N*) and maximum likelihood estimates (with asymptotically normal 95% confidence intervals (CI) in parentheses) of the L_{50} from logistic regression or stanza changing length (SCL) and the relative pros and cons of each method

Method	Sex	*N*	L50 or SCL (cm)	Pros	Cons
GSI	Male Female	425 460	14.27 (13.98–14.57) 14.47 (14.15–14.8)	Good precision/accuracy (based on CI) Not biased by individual perception	High financial costs Medium sampling and processing time High fish mortality
Macroscopic	Male Female	363 409	13.48 (12.95–14.01) 14.01 (13.58–14.43)	Good precision/accuracy (based on CI)	High financial costs Needs qualified personnel Biased by individual perception High sampling and processing time High fish mortality
LWR	Male Female	261 292	15.48 (13.41–17.55) 15.82 (13.34–18.3)	Low financial costs Not biased by individual perception Low fish mortality Low processing time	Can be a biased or imprecise indicator of maturity (based on CI)

3.1 | Logistic Model: Macroscopic gonadal identification and GSI approach

In both analyses, females presented a slightly higher L_{50} than males, but the estimates using the GSI were higher for both genders considering the two approaches (Figure 1, Table 1).

3.2 | Length–Weight Relationship (LWR)

According to the analysis of proportional residues of the Huxley LWR, the monophasic allometric growth model was inadequate for both females ($r^2 = 0.006$; $p = 0.043$) and males ($r^2 = 0.007$; $p = 0.046$) and a trend in biased residuals emerged along total length (Figure 2). This

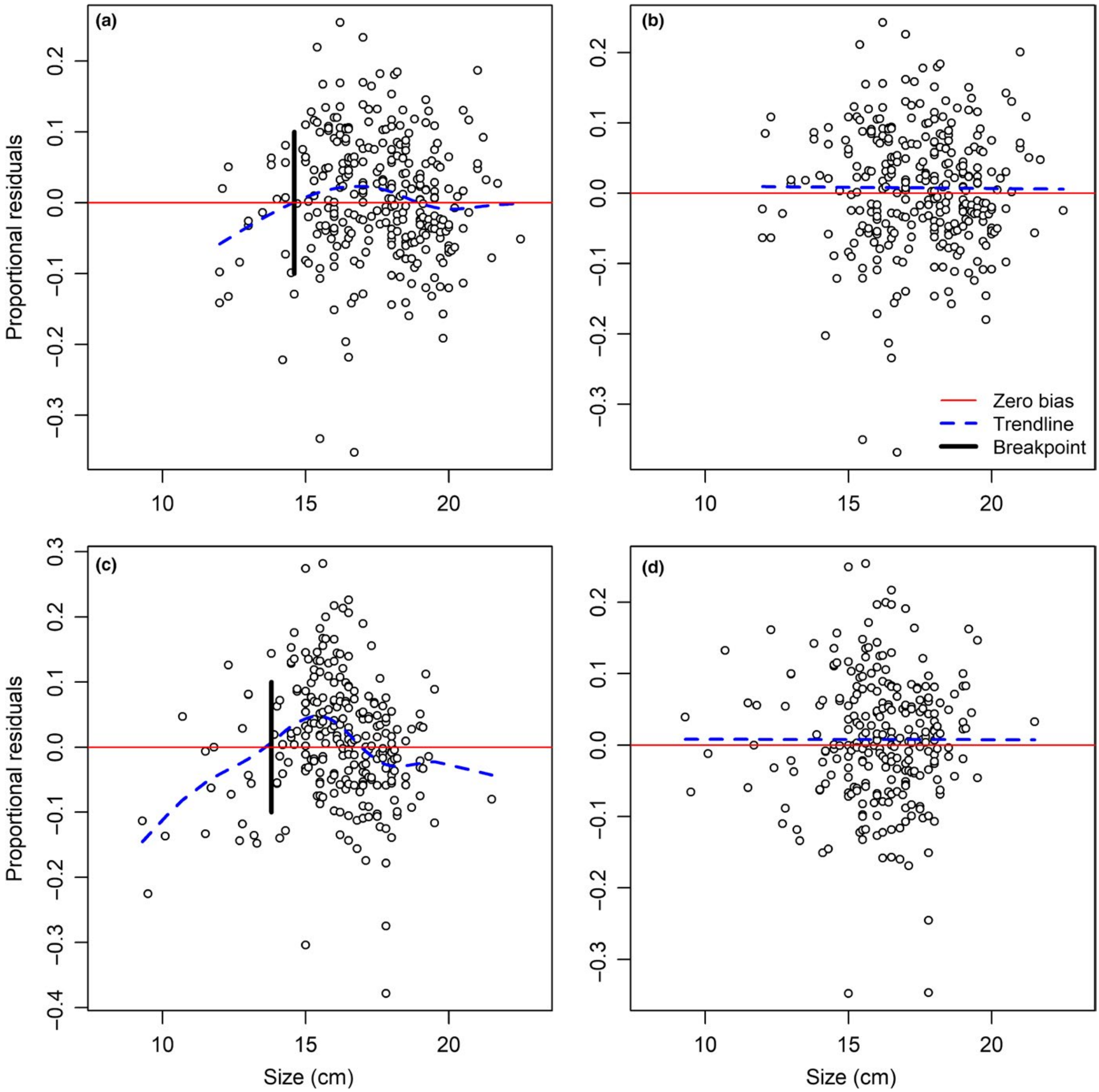

FIGURE 2 Trends in the proportional residuals distribution across size for male (lower panels) and female (upper panels) individuals presuming monophasic (left panels) and polyphasic (right panels) growth in *Cyphocharax abramoides* collected in lower Anapu River, Brazil

bias concentrated with negative residuals (and consequently overestimation of the weight by the monophasic equation) until about 14 cm where an apparent shift in allometry occurred suggesting potential for polyphasic growth among *C. abramoides* individuals (Figure 2).

The polyphasic growth model helped account for the bias across sizes and improved LWR estimation while detecting the apparent shift in allometry occurring around 14 cm (Figure 2). The estimated parameters for the polyphasic growth females showed a change in growth rate at 15.82 cm with positive allometric growth in the juvenile phase (*b* = 3.30) and negative allometric growth in the adult phase (*b* = 2.85). For male *C. abramoides*, a change in growth rate was observed at 15.48 cm with positive allometric growth in the juvenile phase (*b* = 3.43) and negative allometric growth in the adult phase (*b* = 2.60, Table 1).

4 | DISCUSSION

Choosing the most appropriate technique to estimate size at first maturity (L_{50}) in fishes is a hard task. This is so because it depends on operational and biological issues. From an operational perspective, it is necessary to evaluate project needs, as well as available resources and qualified personnel. From a biological perspective, accuracy will be variable due to the precision in data acquirement and due to spatiotemporal variation in reproductive traits.

In this study, we estimated the L_{50} of the detritivorous species *Cyphocharax abramoides* using three different methods—A macroscopic maturational stage classification, the Gonadosomatic Index (GSI) approach and the Length–Weight relationship for the Stanza Changing Length (SCL). The macroscopic identification method and the GSI approach exhibited good precision/accuracy. Nonetheless, their high operational requirements highlighted LWR as a good choice to estimate the size at first maturity despite its less precise estimation. LWR exhibits less operational requirements in contrast with its more precise counterparts.

The GSI considers a proportion of the gonadal and total body weight and is generally used to determine the breeding season of a species (e.g., Oliveira, Fontoura, & Montag, 2011). However, it can also be used as proxy of sexual maturity and therefore another way to determine maturational stages by length class (Fontoura et al., 2009). In this study, the GSI approach presented a good estimate with the lowest 95% confidence intervals indicating the efficiency of the model. It should be emphasised that this analysis should consider the type of spawning of the target species and only use data collected during the reproductive period; otherwise, it will introduce nonreproductive specimens into the sampled population leading to an artificially low GSI_{max} values, inducing an important error in the estimate of the L_{50} (Marques et al., 2007).

Finally, when considering the SCL as a predictor of $L_{50,}$ as proposed by Fontoura et al. (2010), we found that the obtained L_{50} were higher and less precise but with 95% confidence intervals that covered the L_{50} estimates from both macroscopic identification and GSI approach for both genders. This method also highlights another use for the allometric factor obtained in the LWR. Allometric changes in growth rates are well known for fish and starts at pre-larval and larval stages (Gisbert, 1999; Osse & van den Boogaart, 1995) and endures until adulthood due to shifts in energy intake and allocation, according to reproductive investment reflecting environmental and food availability changes (Boukal, Dieckmann, Enberg, Heino, & Jørgensen, 2014; Quince et al., 2008a). Unlike monophasic growth models that assume constant traits through individual lifespans, polyphasic models allow traits to vary in different growth phases most often caused by a shift in reproductive investment (Wilson et al., 2018). Hence, the reproductive strategy of studied species should be considered when using polyphasic models because reproductive investment could start a year before first spawning, as observed in longer-lived fishes. Polyphasic models can be readily incorporated into future studies, but the specific model structure chosen (e.g., change in growth at size at maturity, migration or habitat use) will depend on the ecological hypothesis explaining the change in growth (Wilson et al., 2018).

Cyphocharax abramoides exhibited positive allometry in the first phase and negative allometry in the second phase for both genders, but females exhibited higher allometric coefficients when adults than males. Similar results were found by Fontoura et al. (2010) for characids, and these findings may indicate that fish species need to become larger faster prior to maturity, and that females gain more weight than males after maturity due to higher energy demands for egg development. Also, fish that occupy a low position in the food chain do not need to grow as fast after reaching majority as large predators that may require to be sufficiently larger than their prey (Pawar, Dell, & Savage, 2012).

Results confirm the potential for LWR to estimate size at first maturity in fishes exhibiting polyphasic growth. Thus, we highlight the potential of the LWR for the study of fish stocks as a route for a more practical L_{50} assessment, since it only uses length and weight information of the individuals. Among the three different techniques performed in this study, we concluded that LWR is an affordable and less lethal manner to estimate the size at maturity, and still more studies are necessary to evaluate and improve the accuracy of the tool and to biologically relate it to sexual maturity.

ACKNOWLEDGEMENTS

All authors agree with the submission and have no conflict of interest to declare and thank three anonymous reviewers for their helpful comments. This manuscript was partially funded by the "Programa de Pesquisa em Biodiversidade" (PPBio). DTH is funded by CNPq and received a one-year scholarship from CAPES (PDSE). BES is funded by CNPq. LFAM receives a productivity grant from CNPq (305017/2016-0) and a senior internship scholarship from Texas A&M University (process 88881.119097/2016-1). KLW is funded by a Vanier Canada graduate scholarship and a Killam pre-doctoral scholarship.

CONFLICT OF INTEREST

The authors have no conflict of interests to declare.

AUTHORS' CONTRIBUTION

DTH, conceived and designed the investigation; DTH, performed field and/or laboratory work; DTH, BES, KLW and RDOR, analysed the data; DTH, BES, KLW and LFAM, contributed materials, reagents, and/or analysis tools; DTH, BES, KLW and LFAM, wrote the paper.

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How to cite this article: Hashiguti DT, Soares BE, Wilson KL, Oliveira-Raiol RD, Montag LFDA. Comparing three methods to estimate the average size at first maturity: A case study on a Curimatid exhibiting polyphasic growth. *Ecol Freshw Fish*. 2018;00:1–8. https://doi.org/10.1111/eff.12451