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Application of Stock Density Indices as a Tool for Broad-Scale Population Assessment for Four Cyprinid Species in Central Italy

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ARTICLE

Application of Stock Density Indices as a Tool for Broad-Scale Population Assessment for Four Cyprinid Species in Central Italy

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Abstract
Stock-density indices are standardized methods for analyzing length-frequency data that quantify the length structure of a fish population into length categories that are of interest to recreational anglers. We adapted North American relative stock density (RSD) and proportional stock density (PSD) indices to four Italian endemic or native cyprinid species by means of two methods. In accordance with the traditional approach, the benchmarks of length categories were established according to Gabelhouse's percentage classification and calculated on the basis of the largest individual in the data set for each species. In the second method, asymptotic length and size at maturity were used to define the length categories for index calculation. Both methods were tested on length-frequency data from fish collected from sampling sites in the Tiber River basin. The results showed that the traditional approach displayed better applicability, required less sampling effort, and provided a better insight into the length structure of the fish populations studied.

The length structure of a fish population is one of the most commonly used assessment tools in fisheries, as it reflects interactions among the dynamic rates of recruitment, growth, and mortality. Length-frequency distributions are among the oldest methods used to assess length structure in fish populations (Everhart and Youngs 1981). However, these can often be difficult to interpret because there are few standards by which to assess whether the length frequency is optimal or expected for a given situation. Proportional stock density (PSD) and relative stock density (RSD) enable length-frequency data to be estimated numerically and provide appropriate numerical estimations of deviations from natural length structures in fish populations. Such indices are commonly applied in North American fish communities, and yield results that are easy to communicate to fishery managers (Gassner et al. 2003). Proportional stock density is the percentage of "quality-sized" individuals within the total number of "stock-sized" individuals. Stock and quality size designations are based on the response and expectation of anglers; fish of stock length have little recreational value, while quality length is the minimum size that most anglers like to catch.

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Proportional stock density compresses the entire length-frequency distribution into a single number, thereby engendering a probable loss of information (Gabelhouse 1984). To analyze the length structure of a fish population in greater detail, Gabelhouse (1984) added three additional categories: the proportion of "preferred-length" fish, the proportion of "memorable-length" fish, and the proportion of "trophy-length" fish in the population. Gablehouse (1984) called this length categorization system the RSD. The reference values for each length class vary by species and are defined as percentages of the length of the all-tackle world-record fish. Because PSD and RSD do not always reflect population density (Willis et al. 1993), Guy et al. (2007) suggested that proportional stock density be changed to proportional size distribution.

Although both PSD and RSD are frequently used in North America, they are rarely used in Europe. The first attempt to apply North American PSD and RSD indices in Europe was made by Gassner et al. (2003) to assess the ecological integrity of two Austrian prealpine lakes according to the European Union's Water Framework Directive (WFD) (European Parliament and Council 2000). In their study, the authors used a different method to determine the thresholds for index calculation, based no longer on the percentage of all-tackle world-record fish, but on two biological parameters: the maximum length recorded for the species examined in the study area and the length at maturity. Zick et al. (2007) applied this method to Arctic Char *Salvelinus alpinus* in Austrian lakes, and Pedicillo et al. (2010) applied it to Brown Trout *Salmo trutta* in central Italian streams, using the asymptotic length (L_∞) of the von Bertalanffy growth model and length at maturity. Volta and Oggioni (2010) did the same in order to develop a multimetric index (Lake Fish Index) to assess the ecological status of Italian lakes in accordance with the WFD. In this case, those authors reported reference values for the stock and quality lengths of the key fish species in Italian lakes. As yet, however, reference values for the length classes of fish species in running waters in Italy are lacking. As these indices may be useful indicators in future assessments of the quality of Italian rivers according to the WFD, the goal of our research was to determine, with regard to the principle native fish species present in the Tiber River basin, the reference values delimiting the length classes used to calculate structure indices by means of the two methods available in the literature: the traditional method of Gabelhouse (1984) adapted to the local conditions of fish species in the Tiber River basin (traditional approach), and that of Gassner et al. (2003) modified by Zick et al. (2007) (biological approach). The fish species considered were the cyprinids Cavedano Chub *Squalius squalus*, Rovella *Rutilus rubilio,* Horse Barbel *Barbus tyberinus*, and Italian Riffle Dace *Telestes muticellus*.

The additional aim was to compare the length categories obtained by the two methods and structural indices computed from the length categories by the two methods for sample sites across the Tiber River basin.

METHODS

The study area consisted of the watercourses in the upper and middle portions of Tiber River from its source to its confluence with the Treja River (Figure 1). The Tiber River, the third-longest river in Italy, originates on Mount Fumaiolo (about 1,270 m above sea level) and is 406 km long. Its basin, the second-largest Italian river catchment, covers more than 17,000 km^2 and has an average elevation of 524 m. The area investigated, corresponding to a surface area of 9,832 km^2 (57% of the total drainage area of the Tiber River), included numerous tributaries, the most important ones being the Nestore River (watershed = 1,033 km^2), the Paglia River (1,338 km^2), the Chiascio River (5,963 km^2), and the Nera River (4,280 km^2) (Mearelli et al. 1994). More detailed information on the characteristics of the Tiber River basin and its fish populations is available in Lorenzoni et al. (2006) and Pedicillo et al. (2010). The field studies were carried out between 1992 and 2008; a total of 70 streams and rivers were included in the study for a total of 113–253 sampling stations, depending on the species. Sampling was performed by means of

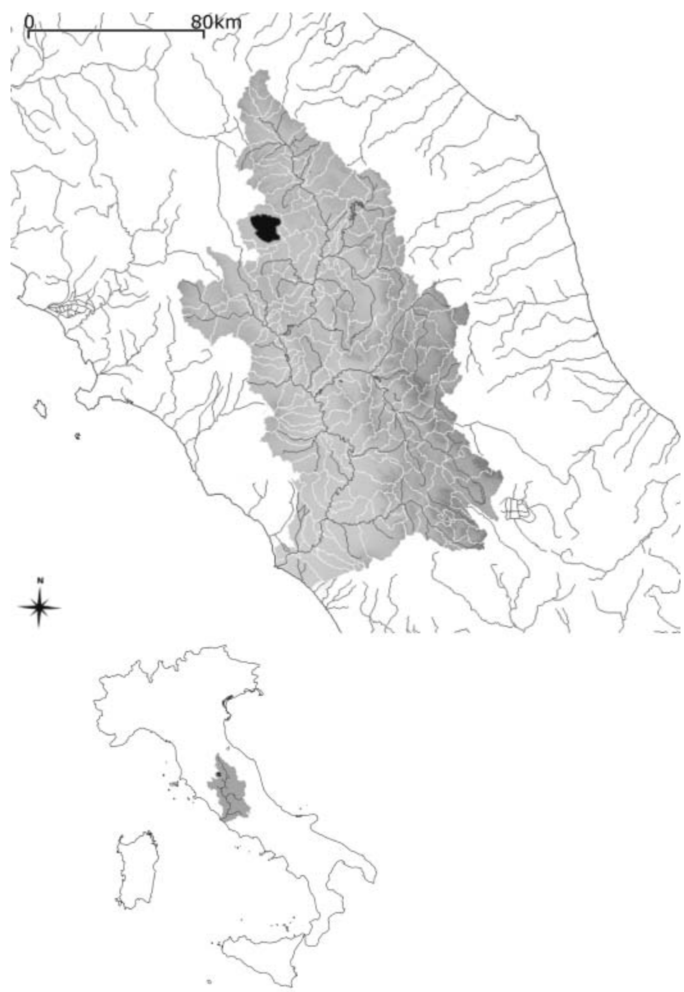

FIGURE 1. Study area (in grey) shown in the River Tiber basin in central Italy.

electrofishing with electric stunning devices of different powers, according to the features of the watercourse involved. All specimens caught were identified by species, counted, and measured (total length [TL] ± 1 mm) (Anderson and Neumann 1996), and scale samples for age determination were taken from the body area, as described by DeVries and Frie (1996). When large numbers of specimens were sampled, scales were only collected from a subsample for each 10-mm length increment. Age determination by means of scale analysis was confirmed and integrated by applying Petersen's length-frequency method (Bagenal and Tesch 1978).

To assess the minimum lengths for the size categories for the RSD index calculation, two different methods were used. For a given species, length classes are traditionally defined as percentage lengths of the all-tackle world-record fish (Gabelhouse 1984). In accordance with this method, length-categorization standards for stock, quality, preferred, memorable, and trophy length were established by adopting the percentages suggested by Gabelhouse (1984) but adapted to the local conditions. Indeed, because there are not official world records for the fish species considered in this study, we used the largest fish in our data set (TL_{max}) for each species (traditional approach). The length of largest fish in the sample is typically used for species lacking an official world record (Milewski and Brown 1994; Zick et al. 2007; Pedicillo et al. 2010). The minimum lengths of the categories were established by using the arithmetic mean of each length class, rounded to the nearest 5-mm increment. For example, to select the minimum stock length for Horse Barbel, a length-range equivalent to 20–26% of the largest fish in our data set ($TL_{max} = 502$ mm) was chosen; the corresponding length class was 101–131 mm and the proposed stock length was 120 mm.

With regard to the second method (biological approach), in accordance with Gassner et al. (2003), two thresholds were defined for each species considered: the asymptotic length (L_∞) of the von Bertalanffy (1938) growth function and the length at maturity (L_m).

The asymptotic length was expressed as the mean value estimated in the study area; the von Bertalanffy growth parameters were calculated by using the mean length at age,

$$L_t = L_\infty \left\{1 - e^{[-k(t-t_0)]}\right\},$$

where L_t is the theoretical length at age t, k is the rate at which the asymptotic length is approached, and t_0 is the theoretical age (in years) at which the length of the specimens is zero. Some populations do not have asymptotic growth trajectories; therefore, the L_∞ and k parameters calculated by means of this equation may be unrealistic (Živkov et al. 1999). To ensure that our analyses were not affected by such problems, we excluded populations with L_∞ more than 50% greater than the maximum length observed in each population (Pedicillo et al. 2010).

The length at maturity was calculated from the mean L_∞ by applying the empirical equation $\log_{10}L_m = 0.8979 \log_{10}L_\infty - 0.0782$ (Froese and Binohlan 2000).

On the basis of these thresholds, in accordance with the biological approach, the length classes were defined as follows (Gassner et al. 2003; Zick et al. 2007; Pedicillo et al. 2010; Volta 2010; Volta and Oggioni 2010): Stock (S) = $Q - [(T - Q)/3]$, Quality (Q) = L_m, Preferred (P) = $Q + [(T - Q)/3]$, Memorable (M) = $Q\{[(T - Q)/3]2\}$ and Trophy (T) = 80% of the L_∞.

Both methods were tested on the length classes for the PSD [(number of fish ≥ minimum quality length / number of fish ≥ minimum stock length) × 100] by analyzing their applicability, the number of specimens needed in order to obtain reliable estimates of PSD, and the efficacy of this index in picking out the differences in the length structure of different samples.

The applicability of the two methods was evaluated with regard to each species by comparing the percentages of sampling sites for which it was not possible to determine PSD because the length of all specimens was below stock length.

According to Willis and Scalet (1989) a fish population with a high PSD consists mainly of large individuals, whereas a population with a low PSD consists mainly of small individuals. The ability of PSD to vary as a function of the fish mean length (Miranda 2007) was used to assess the efficacy of the two methods proposed in order to highlight the differences in length composition of the samples analyzed. For each species, the PSD values calculated by the two methods were plotted as a function of the mean length of specimens from each sampling site, and the regression parameters were examined (Pedicillo et al. 2010). Our basic assumption was the relationship between percentage of large fish in a sample and the corresponding mean length: the higher the percentage of large fish (and hence the PSD) is, the higher the mean length of fish in the sample will be. An ANCOVA was used to compare the two relationships (traditional approach versus biological approach) for each species using mean TL as continuous predictor and the methods (traditional and biological approaches) as factor. The validity conditions for the ANCOVA have been checked by means of the Levene's test for homogeneity of variances.

To demonstrate the utility of stock density indices for assessment of fish-size structure and to examine the sensitivity and applicability of the methods to variations in length structure, the stock and quality lengths for Cavedano Chub were applied to the sampling data from a Chiascio River site. This population was sampled during the autumn in two different years, 1999 and 2005. Indeed, about this time changes in the quality habitat of the sample site occurred and at the same time the population underwent changes in size structure.

Confidence intervals (CIs) for PSD were calculated on the basis of sample size of stock-length fish using normal approximation (Gustafson 1988). The mean values of PSD calculated for each species by means of the two methods were compared using the Mann-Whitney U-test. The chi-square test for

TABLE 1. Total length (mm) descriptive statistics for fish species collected in overall sample.

		TL values (mm)							
Species	Sampling sites (n)	Specimens (N)	Mean	Median	Minimum	Maximum	25th percentile	75th percentile	SD
Horse Barbel	211	13,708	157.9	162.0	20.0	502.0	100.0	205.0	69.3
Cavedano Chub	253	24,873	140.3	120.0	15.0	470.0	75.0	191.0	79.3
Rovella	207	28,008	77.8	75.0	15.0	208.0	60.0	94.0	26.9
Italian Riffle Dace	113	12,996	76.9	75.0	20.0	190.0	52.0	98.0	28.1

differences in probabilities according to Conover (1980) was used to analyze the differences in PSD values from two sampling sites. The thresholds calculated for preferred, memorable, and trophy classes are not discussed here. They have, nevertheless, been provided, as they may contribute to the development and dessemination of these indices.

RESULTS

The length composition of the total sample for each species considered is reported in Table 1. The maximum TL observed for each species was used to define the thresholds for the stock-density index calculation according to the traditional approach (Table 2). Based on the biological approach, the von Bertalanffy growth function parameters of the populations used to generate the mean asymptotic length and the length at maturity for each species can be found in Appendix 1 (available online at https://bio.unipg.it/download/PSD/Appendix1), while the length-categorization system is summarized in Table 3.

The two methods produced different results. Specifically, the proposed stock length according to the traditional approach was considerably lower than that calculated by the biological approach in all species selected. This aspect influences the possibility of applying these structure indices. Indeed, the higher the value of the stock length is in relation to the lengths typical of the species, the greater the probability will be that the

TABLE 2. Traditional approach: classification of the length classes and minimum thresholds for index calculation. The thresholds were established by adopting the percentages suggested by Gabelhouse (1984) and calculated on the basis of the largest fish in the data set (TL_{max}).

Species	Size category	Percent (%) of the maximum length (Gabelhouse 1984)	Length classes (mm) based on maximum length	Minimum length (mm) for size category
Horse Barbel (TL_{max} = 502 mm)	Stock (*S*)	20–26	100–131	120
	Quality (*Q*)	36–41	181–206	190
	Preferred (*P*)	45–55	226–276	250
	Memorable (*M*)	59–64	296–321	310
	Trophy (*T*)	74–80	371–402	390
Cavedano Chub (TL_{max} = 470 mm)	Stock (*S*)	20–26	94–122	110
	Quality (*Q*)	36–41	169–193	180
	Preferred (*P*)	45–55	211–258	230
	Memorable (*M*)	59–64	273–301	290
	Trophy (*T*)	74–80	348–376	360
Rovella (TL_{max} = 208 mm)	Stock (*S*)	20–26	42–54	50
	Quality (*Q*)	36–41	75–85	80
	Preferred (*P*)	45–55	94–114	100
	Memorable (*M*)	59–64	123–133	130
	Trophy (*T*)	74–80	154–166	160
Italian Riffle Dace (TL_{max} = 190 mm)	Stock (*S*)	20–26	38–4.9	40
	Quality (*Q*)	36–41	68–78	70
	Preferred (*P*)	45–55	86–105	90
	Memorable (*M*)	59–64	112–122	120
	Trophy (*T*)	74–80	141–152	150

TABLE 3. Biological approach: classification of the length classes and minimum thresholds for index calculation. The thresholds were established on the basis of the asymptotic length (L_∞) of the von Bertalanffy (1938) growth function and the length at maturity (L_m).

Species	Size category	Equation for the size category calculation	Minimum length (mm) for size category
Horse Barbel	Stock (*S*)	$S = Q - [(T - Q)/3]$	220
$L_m = 257.3$ mm	Quality (*Q*)	$Q = 257.3$	260
$L_\infty = 454.8$ mm	Preferred (*P*)	$P = Q + [(T - Q)/3]$	290
	Memorable (*M*)	$M = Q + \{[(T - Q)/3] \times 2\}$	330
	Trophy (*T*)	$T = 454.8 \times 0.8$	360
Cavedano Chub	Stock (*S*)	$S = Q - [(T - Q)/3]$	250
$L_m = 290.8$ mm	Quality (*Q*)	$Q = 290.8$	290
$L_\infty = 521.3$ mm	Preferred (*P*)	$P = Q + [(T - Q)/3]$	330
	Memorable (*M*)	$M = Q + \{[(T - Q)/3] \times 2\}$	370
	Trophy (*T*)	$T = 521.3 \times 0.8$	420
Rovella	Stock (*S*)	$S = Q - [(T - Q)/3]$	120
$L_m = 137.6$	Quality (*Q*)	$Q = 137.6$	140
$L_\infty = 226.6$	Preferred (*P*)	$P = Q + [(T - Q)/3]$	150
	Memorable (*M*)	$M = Q + \{[(T - Q)/3] \times 2\}$	170
	Trophy (*T*)	$T = 226.6 \times 0.8$	180
Italian Riffle Dace	Stock (*S*)	$S = Q - [(T - Q)/3]$	110
$L_m = 137.6$	Quality (*Q*)	$Q = 137.6$	130
$L_\infty = 226.6$	Preferred (*P*)	$P = Q + [(T - Q)/3]$	140
	Memorable (*M*)	$M = Q + \{[(T - Q)/3] \times 2\}$	150
	Trophy (*T*)	$T = 226.6 \times 0.8$	160

indices cannot be calculated for a given population, since all the specimens caught may be smaller than the stock length. On the basis of stock lengths calculated by the traditional approach for Horse Barbel, only in four instances (2%) out of a total of 211 sampling sites investigated were all specimens below this threshold (Table 4); for the other species investigated, none of the specimens caught at any of the sites were below the stock length. By contrast, when the biological approach was applied, the number of sampling sites at which all specimens were below the stock length was far higher, varying from 8% (9 out of 113) for Italian Riffle Dace to 14% for Horse Barbel (30 out of 211) and Rovella (30 out of 207).

TABLE 4. Percentage of sampling sites in which it was not possible to calculate the proportional stock density (PSD) because all specimens were smaller than stock length. The number of sampling sites (x) out of the total (y) is reported as x/y in parentheses.

Species	Traditional approach	Biological approach
Horse Barbel	2% (4/211)	14% (30/211)
Cavedano Chub	0% (0/253)	12% (30/253)
Rovella	0% (0/207)	14% (30/207)
Italian Riffle Dace	0% (0/113)	8% (9/113)

The stock and quality length values calculated by means of the two methods were applied to the remaining sampling sites in order to determine the corresponding value of PSD. The comparison of mean PSD values for each species highlights the statistical differences between the two methods (Table 5). Moreover, the percentage of sampling sites with significant differences between PSD values obtained from the two methods varied from 47.4% for Horse Barbel and Cavedano Chub, to 79.0%, for Italian Riffle Dace. In these sampling sites in which the PSD determined by the traditional approach was significantly higher than that determined by the biological approach the percentage was very high for each species, ranging from 89.0%, for Horse Barbel up to 100.0%, for Rovella and Italian Riffle Dace.

The relationship between the PSD and mean fish length from each sampling site is shown in Figure 2. The Traditional approach displayed a highly significant positive relationship between PSD and fish length in all selected species (Table 6). When the biological approach was applied, however, no relationship emerged between these two parameters for any of the species analyzed, except for Cavedano Chub. For this species, both methods displayed a highly significant relationship between PSD and length, but the traditional approach yielded a higher correlation coefficient (r), coefficient of determination (r^2), and slope than did the biological approach (Table 6). In each of the species considered, the differences between the two

TABLE 5. Comparison between mean proportional stock density (PSD) values determined by a Mann–Whitney U-test. PSD_T = PSD calculated by the traditional approach, PSD_B = PSD calculated by the biological approach, min = minimum value, max = maximum value. $PSD_T \neq PSD_B$: percentage of sampling sites with significant differences in PSD values obtained from the two methods (comparison performed by chi-square test in sampling sites with both reliable PSD values); $PSD_T > PSD_B$: percentage of sampling sites in which PSD_T is significantly higher than PSD_B. The number of sampling sites (x) out of the total (y) is reported as x/y in parentheses.

Species	Mean PSD_T ± 95% CI (min–max)	Mean PSD_B ± 95% CI (min–max)	Mann–Whitney U-test	$PSD_T \neq PSD_B$ (%) (chi-square test: $P < 0.05$)	$PSD_T > PSD_B$ (%)
Horse Barbel	52.55 ± 3.57 (14–93)	40.09 ± 4.57 (12–77)	$Z = 2.337$ $P = 0.019$	47.4 (18/38)	89.0 (16/18)
Cavedano Chub	49.96 ± 3.33 (2–97)	51.46 ± 4.00 (21–79)	$Z = -0.364$ $P = 0.716$	44.7 (17/38)	94.1 (16/17)
Rovella	51.33 ± 3.29 (4–93)	35.97 ± 3.58 (18–63)	$Z = 4.551$ $P < 0.001$	68.4 (26/38)	100.0 (26/26)
Italian Riffle Dace	58.67 ± 4.06 (10–98)	34.32 ± 5.23 (9–60)	$Z = 5.096$ $P < 0.001$	79.0 (15/19)	100.0 (15/15)

relationships proved to be highly significant as determined from ANCOVA.

The age structure of the Cavedano Chub population sampled in the years 1999 and 2005 in a stretch of the Chiascio River is reported in Figure 3. In 1999 the biological water quality was very poor. Indeed the extended biotic index (EBI; Ghetti 1986), an index used to evaluate the overall water quality based on the composition of the macrobenthic fauna, described the sampling

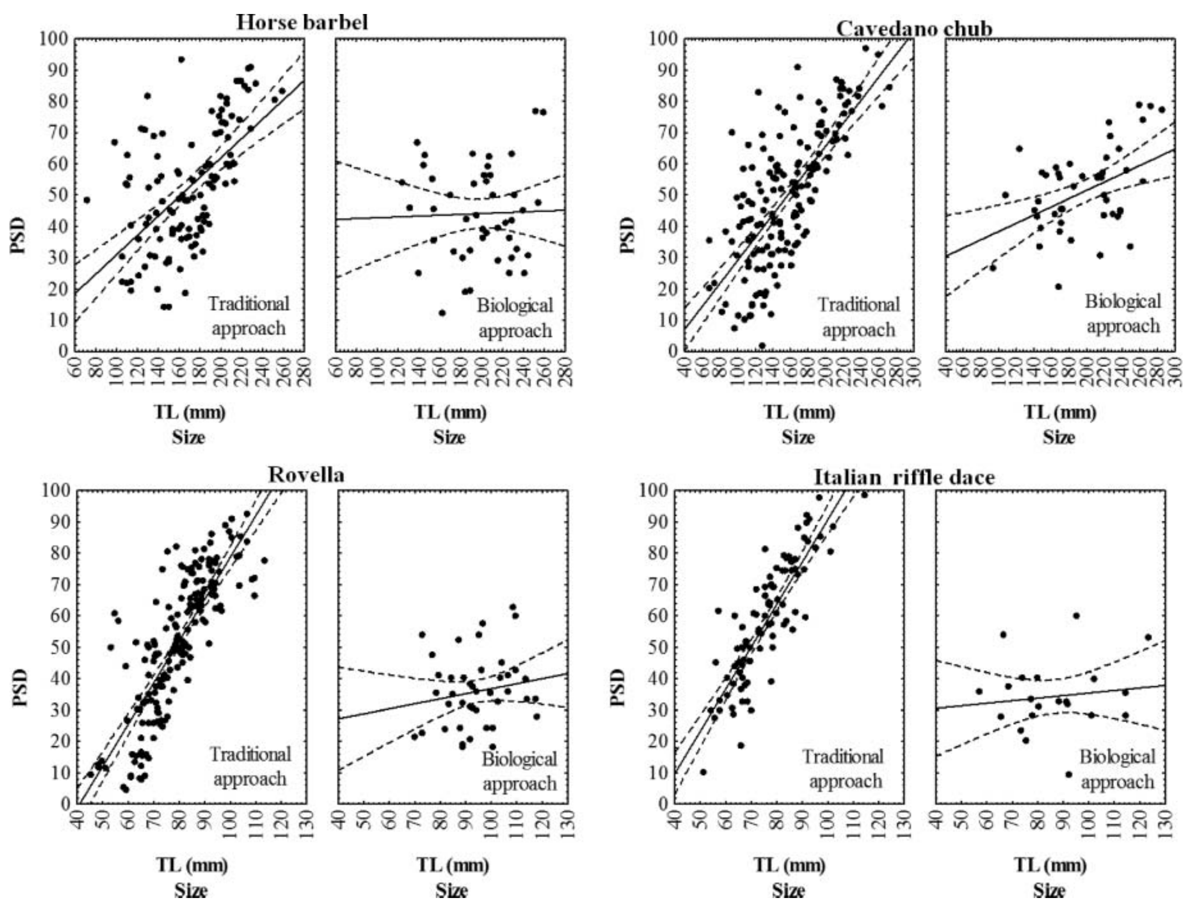

FIGURE 2. Relationship between mean TL of fish samples analyzed and related proportional stock density (PSD) for each species analyzed. Dashed lines denote 0.95 confidence intervals.

TABLE 6. Linear regression analysis (Proportional stock density [PSD] versus mean TL); comparison between the traditional approach and the biological approach was performed by ANCOVA.

	Linear regression results		
Species	Traditional approach	Biological approach	ANCOVA result
Horse Barbel	PSD = −0.07 + 3.10TL $r^2 = 0.337$, $r = 0.581$, $P < 0.001$	PSD = 41.46 + 0.13TL $r^2 = 0.001$, $r = 0.031$, $P = 0.839$	$F = 23.97$, $P < 0.001$
Cavedano Chub	PSD = −7.61 + 3.66TL $r^2 = 0.521$, $r = 0.722$, $P < 0.001$	PSD = 25.14 + 1.32TL $r^2 = 0.203$, $r = 0.451$, $P = 0.002$	$F = 17.71$, $P < 0.001$
Rovella	PSD = −54.42 + 13.31TL $r^2 = 0.654$, $r = 0.809$, $P < 0.001$	PSD = 25.43 + 1.31TL $r^2 = 0.029$, $r = 0.172$, $P = 0.277$	$F = 153.41$, $P < 0.001$
Italian Riffle Dace	PSD = −44.22 + 13.49TL $r^2 = 0.730$, $r = 0.855$, $P < 0.001$	PSD = 27.29 + 0.81TL $r^2 = 0.016$, $r = 0.125$, $P = 0.589$	$F = 104.98$, $P < 0.001$

site as an "environment very polluted" (Lorenzoni et al. 2010). A total of 95 Cavedano Chub specimens were caught and the age structure of the population appeared to be unbalanced; fish varied from age 1+ to age 5+, and the population was dominated by age-2+ fish (70% of the specimens); moreover, the absence of age-0+ fish in the sample indicated that the site was not suitable for the reproduction of the species. Applying the length categorization calculated by means of the traditional approach to the sample, $n = 73$ for the stock-length fish and $n = 20$ for quality-length fish; the related PSD value was 27 (80% CI = ±7.7), demonstrating that there was a shortage of large fish in the population. Based on the biological approach, the stock- and quality-length fish were $n = 9$ and 3, respectively, and consequently the sample size was too small for a reliable PSD estimate (Gustafson 1988). Over time the water quality at the site improved and in 2005 the EBI value described the sampling site as an "environment where signs of pollution can be detected" (Lorenzoni et al. 2012). Moreover the Cavedano Chub population appeared more balanced than it had been in the past: the age structure comprised seven age-classes, from age 0+ to age 6+, with a better distribution of the specimens

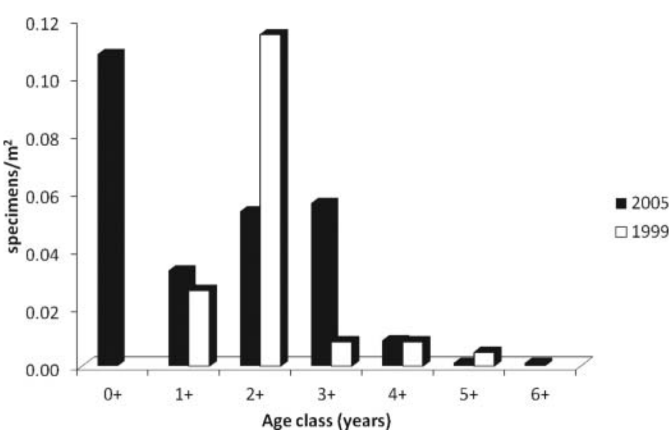

FIGURE 3. Age structures of Cavedano Chub population in Chiascio River for the years 1999 and 2005.

as almost 54% of the specimens were between age 1+ and age 4+. Moreover, fish density increased from 0.162 to 0.261 specimens/m^2 and the number age-0+ specimens was high, demonstrating the suitability of the site for reproduction of the species. Applying the length categorization calculated using the traditional approach the PSD value was 41 (80% CI = ±6.1); $n = 126$ and 51 for stock- and quality-length fish, respectively, out of a total of 208 fish caught. For the biological approach, the sample sizes were insufficient to reliably calculate the PSD (stock-length fish, $n = 4$; quality-length fish, $n = 1$).

DISCUSSION

The assessment of fish populations provides fundamental information for the management of fish species. Indeed, the species considered in this study are endemic in Italy and, except for Cavedano Chub, classified as "Near Threatened" (Rovella, Italian Riffle Dace, and Horse Barbel) according to the International Union for Conservation of Nature criteria (IUCN 2001). Moreover, Cavedano Chub and Horse Barbel are important sport fishes. Readily obtainable indices that reflect the length structure of fish populations and assist in making inferences about population parameters are needed by fisheries managers (Carline et al. 1984) from the point of view of both conservation and sustainable exploitation. Moreover, the establishment of the WFD in 2000 has raised an urgent need to expand our knowledge of aquatic biological communities (including fish) in Italian waterways. The WFD mandates assessment of the ecological status of all European rivers, lakes, and transitional waters on the basis of a wide array of biotic variables, including the composition, abundance, and population structure of fish communities. North American stock density PSD and RSD indices may be useful proxies for determining the population structure of fish, a labor-intensive process that is demanded by the WFD.

The two methods used in our study to calculate the reference values for the length categories in stock-density indices yielded fairly dissimilar thresholds. The values calculated by means of the traditional approach were considerably lower than those calculated by the biological approach in all selected species. The definition of stock length is the minimum length of fish that provides recreational value, while quality length is the minimum size of a fish most anglers would like to catch (Anderson and Neumann 1996). Though Italian Riffle Dace and Rovella are fished in Tiber River basin, no minimum legal limit exists for these species. According to the traditional approach, the minimum stock length for Italian Riffle Dace and Rovella would seem to be of little value when assessing the quality of fishery. In fact fish of this species are small, and in the Tiber River basin they reach 70 mm on average. Hence, the length of 40 mm for Rovella, and 50 mm for Italian Riffle Dace are considered acceptable by local anglers. Moreover, because these species are "Near Threatened," our aim is the establishment of minimum stock and quality lengths that reflect the biology of the species, and which will give the fisheries managers a tool for evaluating the quality of a population structure, not only from the point of view of the fishery but also for the conservation and sustainable exploitation of the species.

According to the biological approach, the length at maturity and the asymptotic length were used to define the specific length classes for PSD and RSD calculation. Generally, the length at which fish of a given population become mature is an important biological parameter for the management (Jennings et al. 1998). Froese and Binohlan (2000) observed that the age at first maturity is a function of size, and they gave an empirical formula to calculate the length at maturity of a given population based on the related asymptotic length, L_∞. Therefore, defining accurate L_∞ values is critical because they are central to the computation of length categories using the biological approach. Indeed, when L_∞ increases the stock and quality lengths also increase. To ensure that our analyses were not biased by unrealistic L_∞ values, we excluded populations with L_∞ greater than 50% higher than the maximum length observed in each population (Pedicillo et al. 2010). Moreover, the coefficient of determination (r^2) may help to evaluate how well the model fits the data. In fact the growth curves used in this study have high r^2 values varying from 0.89 to 1.00, indicating that the models fit the data quite well.

According to the biological approach, minimum quality length is the size at maturity (Pedicillo et al. 2010; Volta 2010; Volta and Oggioni 2010), but the reference values calculated by this method seem to be too high with regard to the local conditions for all species analyzed (Bianco and Santoro 2004; Pompei et al. 2011). These results are in agreement with those of Pedicillo et al. (2010) for Brown Trout. Moreover, for Rovella and Italian Riffle Dace, the length at maturity calculated by means of the empirical formula provided by Froese and Binohlan (2000) proved to be close to the maximum length of the specimens caught in large numbers from the watercourses investigated. Actually the lengths at maturity for some species considered in this study are available in the literature; however, to determine length at maturity, we chose to use the more general formula proposed by Froese and Binohlan (2000). Indeed, the aim of the present study was to select a general method from among those reported in the literature that might be adopted as a model and, if possible, also applied to other fish species, regardless of whether their length at maturity is known. The reference values for the stock length calculated by means of the biological approach also seem to be too high. The value of the stock length is a fundamental aspect in determining the possibility of applying these structure indices. Indeed, the higher the value of the stock length is, the greater the probability will be that the index cannot be calculated, since all the specimens in the sample may be below this threshold. Analysis of the data reported in this study reveals that, for all the species investigated, the thresholds calculated for this length class by means of the biological approach proved to be markedly higher than those determined by means of the traditional approach. Thus, if the reference values calculated by means of the biological approach are used, it is highly likely that, for a given population, the indices cannot be calculated, as the lengths of all the specimens are below the stock length.

When stock and quality lengths calculated by the two methods were used to compute the PSD, they produced different results. The PSD values yielded by the traditional approach were higher than those yielded by the biological approach.

According to Willis and Scalet (1989), low-PSD populations are dominated by small, slow-growing fish with low condition factors. By contrast, high-PSD populations may be less dense, have better growth, and be in better condition. However, other factors can disrupt this relationship (e.g., exploitation) (Willis et al. 1993; Pedicillo et al. 2010). Miranda (2007), in simulated length distributions for three reference species (i.e., Largemouth Bass *Micropterus salmoides*, crappies *Pomoxis* spp. and Bluegill *Lepomis macrochirus*) at three levels of interval annual mortality, found that as mean length decreased, PSD values dropped. Guy and Willis (1990) noted that Largemouth Bass PSD was correlated with mean length of Largemouth Bass in South Dakota ponds. Pedicillo et al. (2010) found a significant relationship between PSD determined by the traditional method and mean length in Brown Trout, while no relation existed between PSD calculated by the biological method and length. Similarly in our study the results of the linear regression between the values of PSD and the mean length of the specimens from the various populations revealed that the traditional approach yielded a significant positive relationship between the sizes of the specimens from the single populations and the related PSD values. However, the r^2 values suggested there is considerable variation in PSD not explained by its relationship with TL. The regression model explaind only about one-third of the variation in PSD for Horse Barbel, while it explained nearly three-quarters of the variation for Italian Riffle Dace. Because Cavedano Chub and Horse Barbel showed greater sizes than Italian Riffle Dace and Rovella, they have a higher size range and consequently higher variance in length distribution. For this reason some barbel or chub populations could have the same PSD value even if they have different mean lengths. This could explain the lower explained percentage by the model for barbel and chub in the PSD–mean length relationship. When the biological approach was used, however, no significant relationship emergeed between these two parameters. The two methods therefore displayed different behaviors according to the lengths of the specimens from the various samples analyzed. Indeed, regression analysis seemed to indicate that the traditional method was more sensitive to changes in the average size of specimens in the samples, and, hence, was better able to highlight the differences in the length-frequency distributions of the populations analyzed (Pedicillo et al. 2010).

Generally, the first attempt to adapt stock-density indices to the local conditions of the Tiber River basin showed that these indices can be useful tools for characterizing fish populations. Indeed, the traditional approach applied to the Cavedano Chub population in the Chiascio River appeared to work quite well, reflecting the temporal variations in age structure of tested population. The objective range for PSD values is typically 30–60 (Penczak et al. 1998) and indicates a balanced fish population. In 1999 the water quality of the sampling site was very poor and the Cavedano Chub population was highly unbalanced. The related PSD value confirmed this condition, being below the objective range, and indicated a population dominated by small fish. Over time the water quality of the sampling site improved and the Cavedano Chub population appeared more balanced with a better distribution of the specimens in the age-classes. Again the PSD value agreed with this change and fells in the objective range, hence indicating a well-structured population. On the contrary, when the stock and quality length calculated by means the biological approach was applied, the sample size became too small for reliable PSD estimates, even if the number of fish caught was quite high in both years. We achieved similar results in a previous study (Pedicillo et al. 2010) in which we tested the methods on Brown Trout length-frequency data from 263 sampling sites in the Tiber River basin. In that study, when the sampling sites were disaggregated according to the various fishing regulations (catch and release, unfished, and fished), the traditional method was better able than the biological approach to reveal the differences in fish-length distribution among the various types of management practices.

Therefore, all analyses seem to show that the traditional method has greater sensitivity and efficacy to highlight the differences in the length-frequency data than does the biological approach and has a greater practical applicability requiring less sampling effort. Hence, this application could contribute to establishing the ecological status of rivers, in accordance with the European Union's WFD (WFD 2000). Moreover the standards provided in this study offer resource managers another tool for identifying and establishing management goals, evaluating management objectives, and providing insight into environmental conditions. Nevertheless fisheries managers should use stock-density indices as only one of their assessment tools. By combining size-structure indices with other tools such as catch per unit effort, fish condition, and growth assessment, population and community analyses will be more reliable.

At present the applicability of the length-category values obtained in this study is limited to the Tiber River basin because they are computed from fish sampled from this area. Important future tasks will be to (1) test the application of indices calculated using the traditional approach on the largest possible number of populations from the Tiber River basin, (2) define the target values for balanced populations, and (3) correlate these indices with other parameters, for example, body condition. Moreover, it will be important to increase our investigations by extending the application of these indices to other fish species.

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