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First information on age and growth of *Padogobius bonelli* (Bonaparte, 1846) outside of its native range: River Aggia (Central Italy)

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Abstract – *Padogobius bonelli* is a goby native to Northern Italy, but several populations are established in Central Italy out of its native range. *P. bonelli* caused negative impact on native fish community but any study on growth in the introduced areas has ever been conducted. In this study age, growth and condition of a non-native population of *P. bonelli* from the River Aggia were investigated. Samples ($n = 422$) were collected monthly from January to December 2012. Age estimation was based on scalimetry and five age classes (0^+–4^+) were observed. Sex ratio was strongly male-biased. Growth in previous years was determined by back-calculation from scale measurements and the occurrence of Lee's phenomenon was tested. Von Bertalanffy growth parameters were estimated (Males: $L_\infty = 8.35$ cm, $k = 0.40$, $\Phi' = 1.44$; Females: $L_\infty = 8.71$, $k = 0.43$, $\Phi' = 1.51$). No differences in growth between sexes were observed, despite sexual dimorphism based on the larger size of males is count as a typical trait of the species. This result could be affected by the reverse Lee's phenomenon that acts only on females, since in the population selectively survived females of larger size. Smaller females would be disadvantaged, especially during the breeding season, as highlighted by the low somatic condition observed in the period April–June.

Key-words: italian gobies / non-native species / growth / condition / back-calculation

Résumé – Premières données sur l'âge et la croissance de *Padogobius bonelli* (Bonaparte, 1846) en dehors de son aire de répartition naturelle : la rivière Aggia (Italie centrale). *Padogobius bonelli* est un gobie originaire d'Italie du Nord, mais plusieurs populations sont établies en Italie centrale hors de son aire de répartition naturelle. *P. bonelli* a causé des impacts négatifs sur la communauté de poissons indigènes, mais aucune étude sur la croissance dans les milieux où il a été introduit n'a jamais été menée. Dans cette étude, l'âge, la croissance et l'état d'une population nonnative de *P. bonelli* de la rivière Aggia ont été étudiés. Les échantillons ($n = 422$) ont été recueillis chaque mois de janvier à décembre 2012. L'estimation de l'âge a été basée sur la scalimétrie et cinq classes d'âge (0^+–4^+) ont été observées. Le sex-ratio était fortement en faveur des mâles. La croissance des années précédentes a été déterminée par rétro-calcul à partir de mesures d'écailles et la présence du phénomène de Lee a été testée. Les paramètres de croissance de Von Bertalanffy ont été estimés (mâles : $L^\infty = 8{,}35$ cm, $k = 0{,}40$, $\Phi' = 1{,}44$; femelles : $L^\infty = 8{,}71$, $k = 0{,}43$, $\Phi' = 1{,}51$). Aucune différence de croissance entre les sexes n'a été observée, en dépit du dimorphisme sexuel et la plus grande taille des mâles rapportée comme un trait typique de l'espèce. Ce résultat pourrait être affecté par le phénomène de Lee inverse qui agit seulement sur les femelles, étant donné que dans la population ont survécu sélectivement les femelles de plus grande taille. Les petites femelles seraient désavantagées, en particulier pendant la saison de reproduction, comme l'a souligné l'état somatique faible observé dans la période avril–juin.

Mots-clés : gobies italiens / espèces non indigènes / croissance / condition / rétro-calcul

1 Introduction

Padogobius bonelli (Bonaparte, 1846) is a bottom-dwelling fish native to Northern Adriatic basin, from Vomano (Italy) to Krka drainages (Croatia) (Bianco and Miller, 1990;

Kottelat and Freyof, 2007). In recent decades *P. bonelli* has been accidentally introduced in several watercourses of Central Italy, together with fish species of fishery interest (Zerunian and Gandolfi, 1986; Bianco and Ketmaier, 2001). The restocking activities with unregulated fish stocks from Northern Italy was a widespread custom that led to the decline

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of native populations of Central Italy due to hybridization, predation and competition (Bianco, 1995). The introduction of *P. bonelli* is a representative example of the negative effects caused by the translocation of species from Northern to Central Italy. *P. bonelli* is counted as one of the main threats to the survival of the congeneric *P. nigricans*, a species endemic to Central Italy, due to: aggressive behaviour and competition for premium reproductive territory (Gandolfi *et al.* 1991; Zerunian, 2004; Mecatti *et al.*, 2010), competition for feeding resources (Pompei *et al.*, 2014) and a higher allocation of energy in reproduction compared to *P. nigricans* (Pompei *et al.*, 2016; Pompei *et al.*, in press.). A full understanding of life history characteristics of alien species is crucial to assess their possible ecological impact on ecosystems, to predict their invasive potential (Ricciardi and Rasmussen, 1998; Guo *et al.*, 2013; Grabowska *et al.*, 2011; Hôrková and Kováè, 2013) and propose effective management strategies (Guo *et al.*, 2013; Yeates *et al.*, 2012; Giannetto *et al.*, 2014). Although the severe ecological impact of *P. bonelli* on native species is wide known, to date, any study on age and growth of non-native populations of *P. bonelli* out of its native range has ever been conducted and basic information on the reproductive biology of this species outside of its native range is only recently available (Pompei *et al.*, in press). Studies conducted on *P. bonelli* from the native areas dealt mainly with its social and reproductive behaviour. For example, extensive researches have been implemented on the production of acoustic signals in males during courtship (Torricelli and Romani, 1986; Torricelli *et al.*, 1986, 1990; Lugli *et al.*, 2004) or during fights for the occupation of territory (Lugli, 1997); other researches focused on the gonadal histology (Cinquetti and Rinaldi, 1987; Cinquetti and Dramis, 2003); still others studies dealt with mating success of males (Bisazza *et al.*, 1989; Marconato *et al.*, 1989). Nevertheless, data on the growth of the native *P. bonelli* populations are rather scarce (Gandolfi *et al.*, 1991) and only general information is available, making it difficult a comparison between native and non-native populations.

The main aim of this study was to provide the first data on age and growth of *P. bonelli* in the non-native area, by analyzing age, growth, sexual size dimorphisms and condition of a population from the River Aggia (Central Italy).

2 Material and methods

2.1 Study area

The River Aggia (43° 24′8.57″N, 12° 12′40.34″E) is a little tributary of the River Tiber, the third-longest (405 km) river in Italy and the second for watershed area (17 375 km^2). The River Aggia is 15.5 km long and it has high-quality waters (Pompei *et al.*, 2014). The banks are continuously covered by dense vegetation and the ground mainly consists of large stones and blocks, interspersed with areas of fine sand (Pompei *et al.*, 2015b). The river's wetted width is quite variable and changes significantly throughout the year, due to the low water flow, which never exceeds 10 L.s^{-1} (Lorenzoni *et al.*, 2010). These characteristics, typical of the Mediterranean climate (with fluctuation between extreme rainfall quantity and extreme drought periods), confer upon the River Aggia a marked torrential regime (Pompei *et al.*, 2016).

P. bonelli was sampled for the first time in the River Tiber basin in 1996, just in the sampling station of the River Aggia investigated in the present study. In 1996 only a few individuals were sampled (Mearelli *et al.*, 1996), since presumably the introduction had been recent; currently, the density of *P. bonelli* in the river sector examined was 1.72 ind.m^{-1}, and the non-native species became much more abundant than the native *P. nigricans* (0.48 ind.m^{-1}) (Pompei *et al.*, 2015a).

2.2 Data collection, processing of the samples and statistical analysis

P. bonelli specimens were collected monthly from January to December 2012 using electrofishing. All *P. bonelli* sampled were immediately anesthetized and then euthanized with an overdose of 2-phenoxyethanol, and were preserved in 4% formaldehyde for the laboratory analysis. Fish were weighed (*W*, nearest 0.1 g) and measured for total length (*TL*, nearest cm) and standard length (*SL*). All specimens were dissected and sex was assessed through macroscopic observation of the gonads; the gonads were removed and weighed to the nearest of 1×10^{-3} g (*W$_g$*).

Chi-squared analysis (χ^2) was used to test the deviation of the sex ratios from parity, both for total sample and separately for each age class.

For each specimen five to ten scales were removed from dorsolateral or ventrolateral rows of the caudal peduncle (Miller, 1975; Pompei *et al.*, 2015b) and preserved in 33% ethanol. Fish scales were observed under a stereo microscope and the number of annuli was counted; two independent age determinations were made by two different operators. An additional age determination was carried out in case of contrasting results. The microscopic scalimetric method was validated by means of length-frequency distribution (Bagenal, 1978).

The relationship between *SL* and *TL* (*SL-TL*) was established for the total sample and separated by sexes using linear regression analysis. Analysis of covariance (ANCOVA) was used to test differences between sexes with *TL* as covariate variable.

The relationship between total length and weight (LWR) was calculated for the total sample and separately for males and females using log transformed data as:

$$W = \log_{10} a + b \log_{10} TL$$

where *a* is the intercept of the regression and *b* the slope or regression coefficient (Froese, 2006). The null hypothesis of isometric growth (H$_0$: *b* = 3) was tested by the *t*-test for both sexes, using $t_s = (b - 3)/S_b$, where t_s is the *t*-test value, *b* is the slope and S_b is the standard error of the slope, for *p* = 0.05 (Sokal and Rohlf, 1987).

Growth in previous years was determined by back-calculation from scale measurements. Back-calculated lengths (BCLs) were estimated for 270 specimens, 124 females and 146 males, analysing 4 scales from each individual.

The distance from the centre of the focus to the anterior edge of the scale along the oblique direction (scale radius: *Sr*)
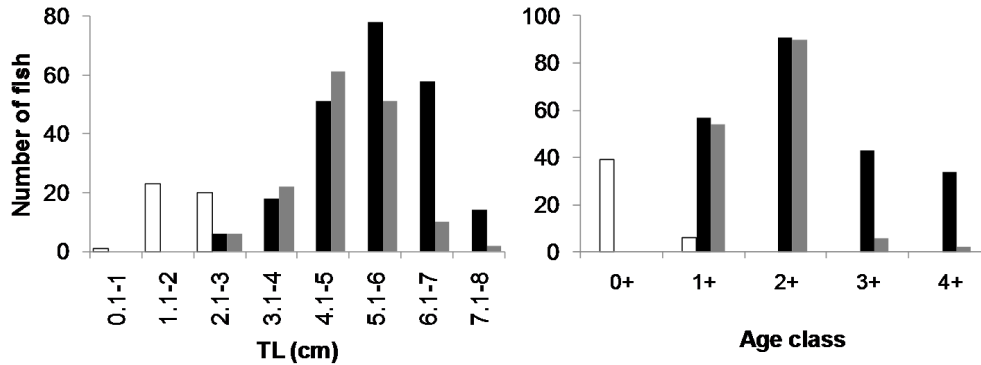
Fig. 1. Total length (a) and age frequency (b) distribution of *P. bonelli* males (black), females (grey) and juveniles (white) from the River Aggia.

and the radius of the annulus (St) in the same direction were measured for each scale (± 0.01 mm) (Bagenal, 1978) using an image-analysis system (IAS 2000). The mean of the measurements of Sr and St was considered in the analysis. The relationship between total body length and scale radius (Sr -TL) was examined separately for males and females through linear regression modelling (Devries and Frie, 1996):

$$TL = a + bSr.$$

Length at age was back-calculated following the Fraser-Lee model (Bagenal and Tesch, 1985) as:

$$Lt = a + StSr^{-1}(TL - a).$$

BCLs at age were determined separately for males and females; the differences between sexes in the BCLs derived from the last annulus were tested by Mann-Whitney U test.

To inspect the occurrence of Lee's phenomenon (Bagenal, 1978), the BCLs reached at the various ages by the specimens at age t were compared with those of the older fish ($t + i$) (Bagenal, 1978) by means of the Mann-Whitney test separately for males and females. Lee's phenomenon is defined as the tendency for BCLs at any age to be smaller, the larger the fish from which they are calculated. In contrast, an inverse Lee's phenomenon is observed when the BCLs are greater, the larger the fish from which they are derived (Bagenal, 1978).

For the description of growth pattern, von Bertalanffy growth model (VBGM) (von Bertalanffy, 1938) was applied:

$$TL_t = L_\infty\{1 - \exp[-k(t - t_0)]\}$$

where TL_t is total length (cm) at age t, L_∞ is theoretical maximum length (cm), k is a constant expressing the rate of approach to L_∞ and t_0 is the theoretical age at which $TL_t = 0$. The values of BCLs from the last annulus were used in the analysis. The index of growth performance (Φ') was calculated following the equation of Pauly and Munro (1984):

$$\Phi' = \log_{10} k + 2 \log_{10} L_\infty$$

where k and L_∞ are the VBGM parameters.

The relative condition factor (Kn) (Le Cren, 1951) was used to evaluate the body condition of the specimens. It was expressed by the formula:

$$Kn = W(aTL^b)^{-1}$$

where a and b are the intercept on the y axis and the coefficient of the LWR calculated on the whole sample, respectively. Somatic relative condition factor (Ks) was also calculated using the somatic weight (Pompei *et al.*, 2012):

$$Ks = (W - W_g)(aTL^b)^{-1}$$

where W is the total weight and Wg is the gonad weight.

Differences between the sexes for both Kn and Ks were tested using the Mann-Whitney U test. The trend of Kn and Ks during the year was examined separately for males and females.

3 Results

The total sample of 422 fish caught in the River Aggia was composed by 227 males, 152 females and 43 juveniles. Males measured from 2.6 to 7.7 cm TL (mean $\pm$ SE = 5.4 $\pm$ 0.07), females from 2.4 to 7.3 cm TL (mean $\pm$ SE = 4.8 $\pm$ 0.07). Females from 4.1 to 6.0 cm TL were the most frequent; in males also individuals of the length class 6.1 to 7.0 cm were abundant (Figure 1a). Five age classes were found, from 0+ to 4+ (Figure 1b).

Sex ratio observed in the total sample (Table 1) was 1.49:1 (M:F) with significant deviations from unity on chi-square analysis (χ^2 = 14.84, $p < 0.01$). Nevertheless the sex ratio changed significantly with age. In the younger age-classes (1^+–2^+) it was well balanced and the deviation from the expected 1:1 ratio was not significant at chi-square test (Table 1), whereas among older specimens, males significantly predominated (Table 1).

No differences emerged between sexes at ANCOVA (F = 1.35, p = 0.25) in the TL - SL regressions. The LWRs (Table 2) resulted to be positive allometric for females, males and total sample, being the values of the regression coefficient (b) significantly greater than the theoretical value of 3 (t-test, $p < 0.05$). Significant differences in the LWR emerged between the two sexes at ANCOVA (F = 5.66; $p < 0.05$).

No differences emerged at ANCOVA in the Sr - TL regressions between sexes (F = 3.86, p = 0.06). Thus the Sr - TL regression calculated for the total sample (Table 2) was used to determine the mean BCLs at age both for males and females (Table 3). Males were larger than females only in the 1^+ age

Table 1. Age composition of *P. bonelli* from the River Aggia expressed as the number (*N*) and percentage (%) of males and females in each age-class. Deviations from unity in the sex ratios (males:females, M:F) were tested by chi-square test (X^2); values of $p < 0.05$ were considered significant.

Age class	Males		Females		M:F	X^2	p
	N	%	N	%			
1+	59	522%	54	478%	1.09:1	0.081	0.776
2+	91	50.3%	90	49.7%	1.01:1	0.006	0.941
3+	43	87.8%	6	12.2%	7.17:1	27.939	<0.01
4+	34	94.4%	2	5.6%	17.0:1	28.444	<0.01
Total	227	59.9%	152	40.1%	1.49:1	14.842	<0.01

Table 2. Standard Length-Total Length (*SL - TL*), Total Length-Weight (LWR) and Total Length-Scale radius (*TL - Sr*) relationships calculated for the total sample, males and females of *P. bonelli* from the River Aggia (r^2 = coefficient of determination; r = correlation coefficient).

			r^2	r	p
SL - TL	Total	$TL = -0.099 + 0.838SL$	0.997	0.999	<0.01
	Males	$TL = -0.155 + 0.847SL$	0.996	0.998	<0.01
	Females	$TL = -0.139 + 0.846SL$	0.992	0.996	<0.01
LWR	Total	$\mathrm{Log}_{10}W = -1.891 + 3.044\log_{10}TL$	0.988	0.994	<0.01
	Males	$\mathrm{Log}_{10}W == -1.941 + 3.112\mathrm{Log}_{10}TL$	0.975	0.987	<0.01
	Females	$\mathrm{Log}_{10}W = -1.979 + 3.169\,\mathrm{Log}_{10}TL$	0.963	0.981	<0.01
TL - Sr	Total	$TL = 1.037 + 34.676Sr$	0.902	0.950	<0.01
	Males	$TL = 0.983 + 34.768Sr$	0.893	0.945	<0.01
	Females	$TL = 0.820 + 36.877Sr$	0.881	0.939	<0.01

Table 3. Mean back calculated lengths (BCLs) in cm ± standard deviation (SD) at successive annuli (L1-L4) in males and females of *P. bonelli* from the River Aggia. Mean BCLs derived from the last annulus are shown in bold.

Males					
Age class	N	L1	L2	L3	L4
1+	27	**3.22 ± 0.37**			
2+	23	3.11 ± 0.31	**4.91 ± 0.47**		
3+	27	3.13 ± 0.30	4.85 ± 0.38	**6.05 ± 0.38**	
4+	9	3.05 ± 0.18	4.83 ± 0.21	6.07 ± 0.15	**6.8 ± 0.17**
Mean ± SD	86	3.15 ± 0.32	4.87 ± 0.39	6.05 ± 0.34	6.8 ± 0.17
Females					
Age class	N	L1	L2	L3	
1+	46	**3.11 ± 0.32**			
2+	36	3.24 ± 0.30	**4.97 ± 0.36**		
3+	3	3.28 ± 0.25	5.40 ± 0.12	**6.42 ± 0.28**	
Mean ± SD	85	3.18 ± 0.32	5.00 ± 0.37	6.42 ± 0.28	

class (Table 3), whereas in the older classes females showed a slightly higher mean BCL (Table 3). Nevertheless, comparison of BCLs between sexes didn't reveal significant differences for any of the age classes at Mann-Whitney *U* test ($p < 0.05$).

Comparisons of mean BCLs of specimens of age *t* and those of *t + i* using the Mann-Whitney *U* test did not reveal any significant differences in males (Table 4), inferring the non-occurrence of Lee's phenomenon. Conversely in females, the BCLs of specimens of age *t + i* were significantly higher than those of age *t* for all the age classes (Table 4), suggesting the occurrence of a reverse Lee's phenomenon.

According to VBGM, females can reach a slightly greater theoretical maximum length ($\pm SE$) ($L_\infty = 8.71 \pm 0.06$ cm) than males ($L_\infty = 8.35 \pm 0.07$ cm) and the growth was also quicker as testified by the values of *k* ($\pm SE$) (females: $k = 0.43 \pm 0.09$; males: $k = 0.40 \pm 0.04$). The index of growth performance (Φ') was greater in females (1.513) than males (1.444).

The mean value of the relative condition factor, Kn ($\pm SE$), was the same for males and females (1.00 ± 0.01; Mann-Whitney *U* test: $Z = 0.17$; $p = 0.86$). Conversely, the relative condition factor calculated with the somatic weight (Ks) was significantly higher in males (0.99 ± 0.01) than females (0.95 ± 0.01) according to the Mann-Whitney *U* test ($Z = 2.99$; $p < 0.01$).

The trend of Kn and Ks throughout the year (Figure 2) was similar between the sexes, with the highest mean values reached in July in males (mean $\pm SE : Kn = 1.12 \pm 0.02$, $Ks = 1.11 \pm 0.02$) and in July and August for Kn and Ks respectively in females ($Kn = 1.08 \pm 0.03$, $Ks = 1.04 \pm 0.03$). The lowest condition was observed in March for both sexes (males: $Kn = 0.91 \pm 0.02$, $Ks = 0.90 \pm 0.02$; females: $Kn = 0.86 \pm 0.03$, $Ks = 0.84 \pm 0.03$). In females the gap between the two parameters is much more evident from April to June, testifying the sharp increase in weight during the reproductive

Table 4. Lee's phenomenon in males and females of *P. bonelli* from the River Aggia: comparison of back-calculated total lengths between specimens at age t and $(t + i)$ for each age class. Results of Mann–Whitney U test are given; values of $p < 0.05$ considered significant.

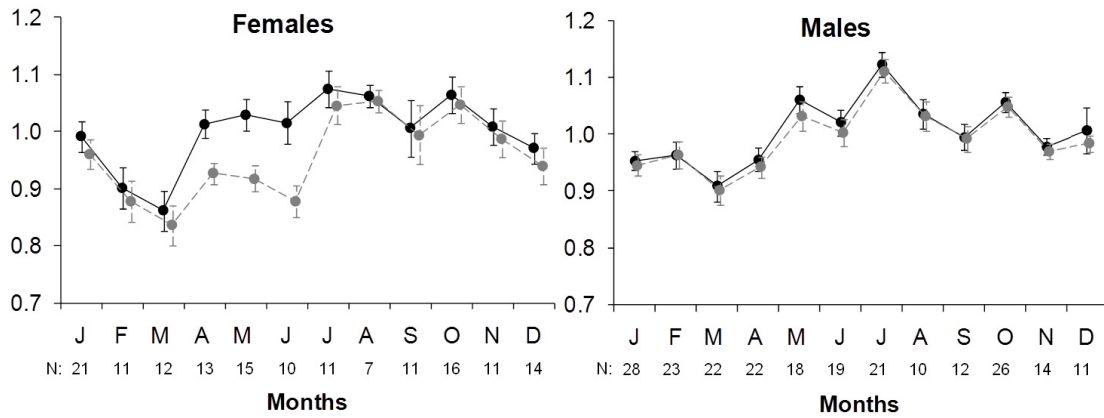|			Age t		Age $(t + i)$	Mann-Whitney U test	
	Age class	N	Mean $TL \pm SE$	N	Mean $TL \pm SE$	Z	p
Males	1+	27	3.22 ± 0.07	59	3.11 ± 0.04	1.279	0.201
	2+	23	4.91 ± 0.10	36	4.85 ± 0.06	0.575	0.565
	3+	27	6.05 ± 0.07	9	6.07 ± 0.05	1.078	0.281
Females	1+	46	3.11 ± 0.05	39	3.25 ± 0.05	2.161	0.031
	2+	36	4.97 ± 0.06	3	5.40 ± 0.07	2.108	0.035

Fig. 2. Monthly changes (mean ± *SE*) of the relative condition factor calculated using total body weight (*Kn*, black circle and solid line) and somatic weight (*Ks*, grey circle and dotted line) in female and male samples of *P. bonelli* from the River Aggia. Sample size (*N*) for each sampling month is indicated.

period due to the development of the ovaries. Nonetheless the mean values of somatic condition factor were particularly low in the period March-June; *Ks* started increasing in July and in August the two parameters coincided again (Figure 2).

4 Discussion

The investigation of growth parameters of non-native *P. bonelli* population from the River Aggia revealed an imbalanced sex ratio towards males. Females predominance is considered a common trait among gobioid fishes (Miller, 1984) and several researches focused on the biology of gobies noted that females outnumber males (Pampoulie *et al.*, 1999; Azevedo and Simas, 2000; Malavasi *et al.*, 2005; Gutowsky and Fox, 2011; Gkenas and Leonardos, 2012; Grul'a *et al,.* 2012). A predominance of females was also observed in a population of *P. bonelli* from the native range for all age classes (Marconato *et al.*, 1989). This sex ratio in favour of females is justified by the reproductive behaviour of the species, since the males are highly territorial and they tend to drive away the conspecific males as a result of space competition (Marconato *et al.*, 1989, Mecatti *et al.*, 2010). Moreover male gobies display high energy investment in nest defence, which could hypothetically lead to depletion and a higher mortality compared to females (Guo *et al.* 2013). Parental care is not only energetically costly but also increases the vulnerability of males to predators (Guo *et al.*, 2013), causing a female predominance. Nevertheless, a male-biased sex ratio is found to be

frequent on goby populations introduced in a new environment (Tomczak and Sapota, 2006; Kornis *et al.*, 2012; Corkum *et al.*, 2004; Young *et al.*, 2010; Gutowsky and Fox, 2011; Roche *et al.*, 2015). A positive relationship between the egg survival rate and the preponderance of males over females has been observed (Kovtun, 1980), since each male will have to guard nests with fewer eggs, resulting in a greater probability of survival of fry: an unbalanced sex ratio towards males would be favoured in populations settled in a new environment (Tomczak and Sapota, 2006; Kornis *et al.*, 2012).

Another interesting result of the study is the absence of sexual size dimorphism in growth, since both maximum body size and theoretical maximum length were similar between the sexes. Moreover, analysing back calculated *TL*, no significant differences between males and females emerged in growth. These results are in contrast to the few available data in the literature for the species: for a population of *P. bonelli* from the native area (Stirone stream), a marked dimorphism in growth was found, with males growing faster and reaching a higher maximum size (Gandolfi *et al.*, 1991). The maximum body size reported in the literature for males ranged between 7.1 and 7.6 (Bisazza *et al.*, 1989; Marconato *et al.*, 1989; Torricelli *et al.*, 1990). On the other hand, a maximum length of 6.4 cm was found for females (Marconato *et al.*, 1989). Sexual dimorphism based on the larger size of males is reported as a typical trait of the species (Bisazza *et al.*, 1989; Gandolfi *et al.*, 1991; Marconato *et al.*, 1989). Among gobies, males are usually bigger than females, as noted in many other researches

(Čápová *et al.*, 2008; Filiz and Toðulga, 2009; Scalici and Gibertini, 2009; Borcherding *et al.*, 2011; Gutowsky and Fox, 2011; Grul'a *et al.*, 2012; Pompei *et al.*, 2015b). Due to male-guarding behaviour, males of several gobies species, such as *P. bonelli,* may be subjected to a strong sexual selection because the larger males will occupy better nests (Bisazza *et al.*, 1989; Marconato *et al.* 1989; Bobbio *et al.*, 1990; Lugli *et al.*, 1992; Scalici and Gibertini, 2009), obtaining a greater chance of attracting a female. In resource-based breeding systems, the availability of resources may be important for both the mating system and intensity of sexual selection (Emlen and Orign, 1977). In nest-guarding species, nest-site abundance has been found to affect mating patterns, and consequently the males' dimensions (Forsgren *et al.*, 1996). Different sexual selection patterns are likely to exist at different stages of invasions of a non-native species. During the early stages of establishment, low density and relatively abundant resources lead to low male-male competition for good-quality nests. In these conditions, no selection for larger males occurs, and consequently the population will not be sexually size dimorphic (Forsgren *et al.*, 1996). Nevertheless, *P. bonelli* from the River Aggia can be considered a long-established population, since it has been present in the river sector investigated for almost 20 years. Moreover, in addition to intraspecific competition, inter-specific competition for breeding sites with *P. nigricans* probably occurred. Although the maximum lengths of *P. bonelli* males from the River Aggia are similar to that found in the literature (7.7 cm), the females were found to be longer. Therefore, it is possible that the absence of sex size dimorphism could be due to other factors, such as a stronger selection for females' fecundity favouring larger female size (Forsgren *et al.*, 1996), in order to maximise the reproductive success in a highly unpredictable environment such as the River Aggia, and facilitating the invasion process (Pompei *et al.*, in press). Another cause for the absence of sexual size dimorphism could be selective mortality affecting the smaller females, as emerged from the analysis of back-calculated lengths, suggesting the presence of a reverse Lee's phenomenon. The interpretation of this phenomenon could possibly be limited by the short lifespan of the species, since comparisons involved only three age classes and the sample is restricted to a single sampling year. Nevertheless the reasons of the occurrence of a reverse Lee's phenomenon are generally found in some kind of pressure, i.e. predation or competition, selectively disadvantaging the specimens that in the first years of life grew slower than the rest of the population and selectively survive specimens of larger dimensions. It remains unclear what factors determine the higher mortality in females. The females are in worse physiological condition than the males, as highlighted by the comparison of the condition factor calculated using the somatic weight (Ks). The variation of Kn and Ks throughout the year showed a sharper decline in the females' condition during winter months than in the males; after March the body condition began to increase in both sexes, but in females the values of Ks remained fairly low throughout the reproductive period (April-June). A previous research reported that *P. bonelli* females from the River Aggia are characterized by a great reproductive investment and the production of a high number of eggs with respect to native populations of Northern Italy (Pompei *et al.*, in press). The huge resources employed in reproduction are put into relation both with the plasticity in life history traits of the invasive species and with a reproductive strategy that enhance a population to survive in instable environment (Pompei *et al.*, in press). This could lead to the hypothesis that the poor physical condition after winter, worsened by the reproductive strain, could be one of the causes of the higher mortality in females than males.

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