

Age and growth of the karyologically identified spined loach *Cobitis taenia* (Teleostei, Cobitidae) from a diploid population

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Abstract. A total of 174 specimens of the spined loach *Cobitis taenia* Linnaeus, 1758, comprising 117 females, 45 males and 12 juveniles, from Lake Klawoj (Poland) were identified as *C. taenia* from their karyotype ($2n = 48$ chromosomes). The overall sex ratio (M:F = 1:2.6) showed that females dominated in the population, but this varied with increasing fish size. Age and growth were determined based on otolith measurements. Standard lengths were back-calculated from the regression of fish standard length on the otolith radius and the growth pattern was described using the von Bertalanffy model separately for males and females, which revealed a strong fit for both females ($L_t = 92.4(1 - \exp(-0.278(t - 0.456)))$; $r^2 = 0.803$) and males ($L_t = 120.6(1 - \exp(-0.227(t - 0.558)))$; $r^2 = 0.772$). Comparison of asymptotic length with maximum observed size of males and females revealed that all values were a bit overestimated (as per the Taylor criteria). The SL – weight equations for males ($W = 0.0003 SL^{3.8377}$; $r^2 = 0.8961$) and females ($W = 0.0039 SL^{3.1683}$; $r^2 = 0.9616$) indicated allometric growth in both sexes, though more so in males than in females.

Key words: otolith, Pisces, von Bertalanffy function, weight-length relationship

Introduction

There are relatively few studies that have examined the age and growth of the spined loach *Cobitis taenia* (Robotham 1981, Boron 1992). Until recently, *C. taenia* was thought to be rare but widespread in Poland, occurring in all types of freshwaters. However, recent cytogenetical investigations (Bohlen & Ráb 2001, Boron 2001) revealed that *C. taenia* is not so common, representing $\approx 13\%$ of all *Cobitis* specimens collected (Boron 2003). Most of the *Cobitis* populations in Poland consist of *C. taenia* and/or *C. elongatoides* and their polyploid hybrids (Boron 2003). However, polyploid fish are not morphologically distinguishable from diploid specimens (Kotusz 2000, Jeleń 2006). All Cobitidae species have been protected under Polish law since 2004. The occurrence of diploid-polyploid *Cobitis* populations in Poland casts doubt on the results of any previous studies that did not provide cytogenetic identification of the specimens examined. According to karyological identification of spined loaches from the Great Ouse River basin (Boron et al. 2003) at least Robotham (1981) studied age and growth of a population of pure diploid *C. taenia*. However, Robotham (1981) studied the species in a small flowing river, though it also occurs in lakes in a different environment. The aim of the study was to provide information on the age and growth of a pure population of *C. taenia* from a lake in Poland and to compare it with the data of Robotham (1981) as well as with data from mixed diploid-polyploid complexes.

Material and Methods

Using a seine net, 174 specimens of *C. taenia*, consisting of only diploid ($2n = 48$) individuals, were collected between April and September, biweekly in 2001 and monthly in 2002, from Lake Klawoj in northern Poland (Borón 1999). Immediately after capture, the fish were anaesthetized, measured for standard length (SL) and total length (TL) to nearest 0.1 mm and weighed (to the nearest 0.1 g.), and dissected to determine sex (117 females, 45 males, 12 juveniles). Each fish was identified according to its karyotype, with chromosome slides made from kidney tissue as per Ráb & Roth (1989) and Borón (1995).

Sex was determined by gonad histology (Juchno et al. 2007) and the presence of the *lamina circularis*. Specimens possessing the *lamina circularis* and those characterized by gonads containing vitellogenic oocytes were considered respectively as sexually mature males and females.

Age was determined from otoliths (Nagięć 1993), which were cleaned, immersed for 24 hours in glycerin (Robotham 1981) and then examined under a Nikon binocular microscope. Distances from the otolith's centre to each annual ring (r_n) and to the otolith's edge (R – total radius) were measured to the nearest 0.05 mm. Back-calculated fish SL for each year of life was by the direct proportion method (Francis 1990, Devries & Frié 1996), using the regression relationship between SL and total otolith radius (R): $SL = 172.84 \times R + 7.674$ ($r^2 = 0.8705$).

Back calculated SL at age were used to estimate the von Bertalanffy growth function (VBGF): $L_t = L_{inf}(1 - \exp(-K(t - t_0)))$ where L_{inf} is the asymptotic length, K is the growth constant and t_0 is the hypothetical time at which the fish started to grow (Ricker 1975). The VBGF parameters were also used to calculate the overall growth index $\phi' = \log_{10}(K) + 2 * \log_{10}(L_{inf})$ (Munro & Pauly 1983). All these parameters (and their standard errors) were calculated using the FiSAT software (Gayani et al. 1994). The SL–weight relationship ($W = aSL^b$) was used to determine whether growth was isometric ($b = 3$) or allometric ($b \neq 3$). To test this pattern, the Student t – test was applied following the log-transformation of data (Zar 1984).

Results and Discussion

Females dominated the Lake Klawoj population of *C. taenia* (M:F = 1:2.6) in both 2001 (1:2.9) and 2002 (1:1.8). This ratio varied according to SL (Fig. 1) and age (Table 1). Robotham (1981) reported a male – female ratio of *C. taenia* of 1:1.3. The higher dominance of females in Lake Klawoj may be caused by higher mortality risk by predation of the smaller males (Bohlen & Ritterbusch 2000). In mixed, diploid-ployploid populations, this ratio was 1:1.75 (Kostrzewa et al. 2003) and 1:22 (Bohlen & Ritterbusch 2000).

Maximum observed SL in males was 72.5 mm (specimen captured in June 2001) and 91.7 mm in females (specimen captured in May 2002). SL at age varied widely due to the combination of specimens collected over a two-year period, which introduced a seasonal growth effect (Rosenberg & Beddington 1988). The equation describing the TL to SL relationship was: $TL = 0.22 + 1.14 \times SL$, $r^2 = 0.9930$ (for females) and $TL = 1.09 + 1.12 \times SL$, $r^2 = 0.9936$ (for males).

C. taenia males from Lake Klawoj were smaller than females (Fig. 1), which is a fixed biological feature of all *Cobitis* species such as *C. paludica* (Przybylski & Valladolid 2000), *C. elongatoides* (Erős 2000), and *C. narentana* (Schneider

Table 1. Number of fish (n), proportion of total sample (%), standard length (SL in mm, with S.E.) and weight (W in g) of males and females by age group for *C. taenia* from Lake Klawój (Poland) in 2001 and 2002.

Age Group	Males				Females			
	n	%	mean SL min – max S.E.	mean W min – max S.E.	n	%	mean SL min – max S.E.	mean W min – max S.E.
1+	0	0	-	-	13	11.1	45.5 33.2–56.1 7.33	0.8 0.3–1.2 0.30
2+	12	26.7	52.9 49.6–56.3 2.23	1.2 0.9–1.6 0.24	13	11.1	59.5 41.4–66.6 7.34	1.7 0.6–3.1 0.69
3+	28	62.2	58.4 53.4–63.4 3.32	1.7 1.3–3.4 0.47	36	30.8	74.6 58.9–85.8 6.82	3.5 1.5–5.7 1.05
4+	5	11.1	69.6 65.3–72.5 2.70	3.7 2.4–4.9 0.97	52	44.4	81.3 70.3–88.5 4.25	4.6 2.8–7.3 0.87
5+	0	0	-	-	3	2.6	88.1 85.9–91.7 3.17	6.2 5.4–7.1 0.84

et al. 2000, Zanello et al. 2003), and was also found by Robotham (1981) in *C. taenia* from the Great Ouse River. Females achieved sexual maturity at about 53.3 mm SL and males at about 40 mm SL (Juchno et al. 2007). The 12 juveniles under the present study ranged in SL from 31.4 to 46.8 mm, and in weight from 0.15 to 0.7 g. The smallest loaches (SL from 31.4 to 36.4 and weight about 0.3 g) collected on 22 June 2001 were likely 1+ specimens, while fish collected one month later, with an SL from 20 to 25 mm, could represent the 0+ cohort. The lengths of the smallest fish, corresponding with fry, attained a length of 15 mm after 24 days given by Robotham (1981).

More than 60% of males in Lake Klawój were age 3+ and \approx 45% of females were age 4+, which is similar to the *C. taenia* population studied by Robotham (1981) and to that of other European species of *Cobitis* (Marconato & Rasotto 1989, Rasotto 1992, Przybylski & Valladolid 2000, Oliva-Paterna et al. 2002, Kostrzewa et al. 2003, Ekmeççi & Erk'akan 2003). Furthermore, the maximum age in Lake Klawój females was age 5+ and age 4+ in males (they should die by the end of their sixth and fifth year of life respectively) (Table 1), which is similar to other *Cobitis* species (Lobon-Cervia & Zabala 1984, Przybylski & Valladolid 2000, Erős 2000, Schneider 2000, Ritterbusch & Bohlen 2000). One exception is *C. taenia* population from the Great Ouse River with the majority of females attained the age of 3+, and the males reached the age of 2+ (Robotham 1981).

Comparison of observed and back-calculated length showed good agreement (Table 2), with annual increments decreasing between age groups in the same magnitude for both sexes and the fastest growth was in the first year of life. The high coefficient of determination values ($r^2 = 0.803$ for females and 0.772 for males) indicate a strong fit of the von Bertalanffy equation to the observed data (Table 3). Although the largest male had 72.5 mm and female had 91.7 mm, the maximum asymptotic length (L_{inf}) was found to be

92.4 and 120.6 mm, respectively. Taking into account the Taylor's (1962) criterion of fitting the growth function to the observed data, the L_{inf} is overestimated for both sexes. Meanwhile, L_{inf} was found to be ≈ 1.3 times higher in females than in males, with similar results reported elsewhere for *C. paludica* (Przybylski & Valladolid 2000) and *C. narentana* (Zanella et al. 2003).

A genetically determined characteristic, such as theoretical maximum length (L_{inf}), can be affected by varying environmental influences (Weatherley & Gill 1987). Therefore the asymptotic SL is considered to reflect growth capacity under particular environmental conditions (Bagenal & Tesch 1978). Growth constant K was only slightly larger in males ($K = 0.278$) than in females ($K = 0.227$), indicating a slightly faster growth rate. Because the inverse relationship between L_{inf} and K is an inherent property of the von Bertalanffy growth function (Moreau et al. 1985), further but reversed differences in growth between sexes are revealed by the index of growth (ϕ'), with a higher value (and faster growth) in females than males (Table 3).

The length-weight equation calculated for males was $W=0.0003SL^{3.8377}$, $r^2 = 0.8961$, while the similar equation for females was $W = 0.0039SL^{3.1683}$, $r^2 = 0.9616$. This indicated that at a similar SL, males achieved a greater weight than females ($t = 3.131$, $df = 158$, $P = 0.0021$). Moreover both sexes grew allometrically with $b>3$ ($t = 4.203$, $df = 43$, $P <0.001$ for males and $t = 2.961$, $df = 115$, $P <0.002$ for females). The similar pattern was also noted for a *C. taenia* population in England (Robotham 1981), a *C. paludica* population in

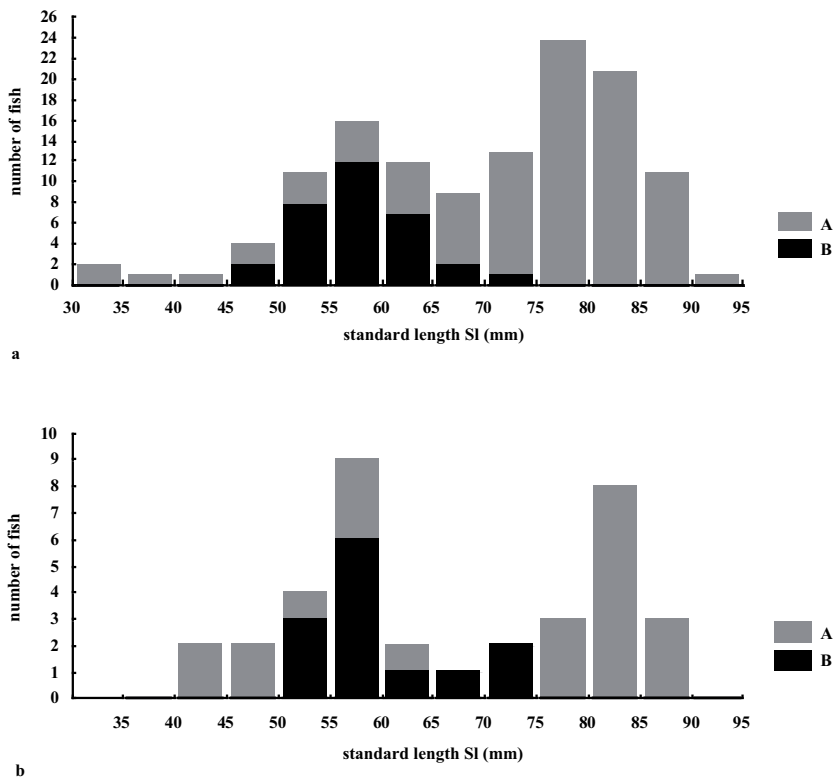


Fig. 1. Standard length-frequency distributions of female (A, grey) and male (B, black) *C. taenia* at capture from Lake Klawój (Poland) in 2001 (a) and 2002 (b).

Table 2. Mean back calculations of standard lengths (SL in mm) obtained from otoliths measurements of *C. taenia* from Lake Klaw6j (Poland) in 2001 and 2002.

Age Group	SL at capture			SL at age				
	n	mean	S.D.	l_1	l_2	l_3	l_4	l_5
Males								
1	-	31.1	5.97					
2	12	45.3	6.94	35.3	48.1			
3	28	55.9	5.89	29.1	43.9	55.6		
4	5	66.4	4.04	31.8	46.2	57.2	66.4	-
Mean length				32.1	46.1	56.4	66.4	
S.D.				3.1	2.1	1.1		
Annual increment					14.0	10.3	10.0	
Females								
1	13	35.1	7.87	38.7				
2	13	51.4	8.53	36.5	52.6			
3	36	66.6	7.40	37.7	55.1	69.9		
4	52	76.7	5.27	31.9	48.5	64.6	76.6	
5	3	82.8	0.75	36.4	50.9	60.7	75.9	82.8
Mean length				36.2	51.8	65.1	76.3	
S.D.				2.6	2.8	4.6	0.5	
Annual increment					15.5	13.3	11.2	6.6

Table 3. Parameters of the von Bertalanffy growth model and their standards errors S.E. for *C. taenia* from Lake Klaw6j (Poland).

Parameters	Males	S.E.	Females	S.E.
L_{inf}	92.4	21.1	120.6	15.6
K	0.278	0.127	0.227	0.056
t_0	-0.456	0.290	-0.558	0.171
ϕ'	3.385	0.655	3.665	0.565
n	130		370	
r^2	0.7718		0.80297	
max. lifespan	4		5	

Iberia (Przybylski & Valladolid 2000) and a *C. narentana* population in Croatia (Zanella et al. 2003). Because the presented result concerns the pure diploid population of *C. taenia*, which is rare in Central Europe, the results could be used as references to compare, for example, how polyploidisation could affect the life history of loaches.

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LITERATURE

- Băcescu M. & Mayer R. 1969: [The *Cobitis* of the Don and Volga]. *Vopr. Ichtiol.* 9: 51–60 (in Russian).
- Bagenal T.B. & Tesch F.W. 1978: Age and growth. In: Bagenal T. B. (ed.), *Methods for Assessment of Fish Production in Fresh Waters*. 3rd edn. *Blackwell, Oxford*: 101–136.
- Bohlen J. & Ráb P. 2001: Species and hybrid richness in spined loaches of the genus *Cobitis* (Teleostei: Cobitidae), with a checklist of European forms and suggestions for conservation. *J. Fish Biol.* 59 (Suppl A): 75–89.
- Bohlen J. & Ritterbusch D. 2000: Which factors affect sex ratio of spined loach (genus *Cobitis*) in Lake Müggelsee? *Environ. Biol. Fish.* 59: 347–352.
- Boroń A. 1992: [Spined loach, *Cobitis taenia* (L.) from Zegrzyński Dam Reservoir]. *Ph.D. thesis, University of Warmia and Mazury in Olsztyn* (in Polish).
- Boroń A. 1995: Chromosome banding studies of spined loach *Cobitis taenia* (L.). *Cytobios* 81: 97–102.
- Boroń A. 1999: Banded karyotype of spined loach *Cobitis taenia* and triploid *Cobitis* from Poland. *Genetica* 3: 293–300.
- Boroń A. 2001: Chromosomal diversity of the fish genus *Cobitis* (Pisces, Cobitidae) distributed in Poland. *Dissertations and monographs UWM* 39: 1–74 (in Polish with English summary).
- Boroń A., Culling M. & Pułym A. 2003. Cytogenetic characteristics of the fish genus *Cobitis* from England. *Folia Biol. (Kraków)* 51 (Suppl.): 13–16.
- Boroń A. 2003: Replication banding patterns in the spined loach, *Cobitis taenia* L. (Pisces, Cobitidae). *Genetica* 119: 51–55.
- Devries D.R. & Frie R.V. 1996: Determination of age and growth. In: Murphy B. R. & Willis D. W. (eds), *Fisheries Techniques. American Fisheries Society, Bethesda, Maryland*: 483–512.
- Ekmekçi F.G. & Erk'akan F. 2003: Preliminary data on growth and reproduction of *Cobitis simplicispina* from Turkey. *Folia Biol. (Kraków)* 51 (Suppl.): 183–186.
- Erős T. 2000: Population biology of *Cobitis elongatoides* in a lowland stream of the Middle Danube (Hungary). *Folia Zool.* 49 (Suppl. 1): 151–157.
- Francis R.I.C.C. 1990: Back-calculation of fish length: a critical review. *J. Fish Biol.* 36: 883–902.
- Gayanilo F.C., Sparre P. & Paul D. 1994: The FAO – ICLARM Stock Assessments Tools (FISAT) user's guide. FAO Computerized Information Series. *Rome: FAO*.
- Jeleń I. 2006: [Diversity of morphological features of the spined loach *Cobitis taenia* Linnaeus, 1758 and natural polyploidy of *Cobitis* (Pisces, Cobitidae)]. *Ph.D. thesis University of Warmia and Mazury in Olsztyn* (in Polish).
- Juchno D., Boroń A. & Gołaszewski J. 2007: Comparative morphology and histology of the ovaries of the spined loach *Cobitis taenia* L. and natural allopolyploids of *Cobitis* (Cobitidae). *J. Fish Biol.* 70: 1392–1411.
- Kotusz J. 2000: Intra- and interpopulation morphological variability in diploid and varied-ploidy *Cobitis* from Poland. *Folia Zool.* 49 (Suppl. 1): 219–226.
- Kostrzewa J., Przybylski M., Marszał L. & Valladolid M. 2003: Growth and reproductive biology of loaches *Cobitis* sp. in Lake Lucień, Poland. *Folia Biol. (Kraków)* 51 (Suppl.): 179–182.
- Lobon-Cervia J. & Zabala A. 1984: Observation on the reproduction of *Cobitis paludica* de Buen, 1930 in the Jarama River. *Cybiu* 8: 63–68.
- Marconato A. & Rasotto M.B. 1989: The biology of a population of spined loach, *Cobitis taenia* L. *Boll. Zool.* 56: 73–80.
- Moreau J., Belaud A., Dauba F. & Nelva A. 1985: A model for rapid growth evaluation in fishes: the case of the Cyprinids of some large French rivers. *Hydrobiologia* 120: 225–227.
- Munro J. L. & Pauly D. 1983: A simple method for comparing the growth of fish and invertebrates. *Fishbyte* 1: 5–6.
- Nagięć M. 1993: Informative function of otoliths. *Biul. Nauk. ART., Olsztyn* (11)/93: 69–80.
- Oliva-Paterna F.J., Torralva M.M. & Fernandez-Delgado C. 2002: Age, growth and reproduction of *Cobitis paludica* in a seasonal stream. *J. Fish. Biol.* 60: 1–16.
- Przybylski M. & Valladolid M. 2000: Age and growth of *Cobitis paludica* in the Lozoya River (Central Spain). *Folia Zool.* 49 (Suppl. 1): 129–143.
- Ráb P. & Roth P. 1989: Chromosome studies in European leuciscine fishes (Pisces, Cyprinidae). Aneuploidy due to a B-chromosome in *Rutilus rutilus*. *Folia Zool.* 38: 333–337.
- Rasotto M.B. 1992: Gonadal differentiation and the mode of sexuality in *Cobitis taenia* (Teleostei; Cobitidae). *Copeia* 1: 223–228.
- Ricker W.E. 1975: Computation and interpretation of biological statistics of fish populations. *Bull. Fish. Res. Bd. Canada* 191: 1–382.

- Ritterbusch D. & Bohlen J. 2000: On the ecology of spined loach in Lake Müggelsee. *Folia Zool.* 49 (Suppl. 1): 187–192.
- Robotham P.W.J. 1981: Age, growth and reproduction of a population of spined loach, *Cobitis taenia* (L.). *Hydrobiologia* 85: 129–136.
- Rosenberg A.A. & Beddington J.R. 1988: Length-based methods of fish stock assessment. In: Gulland J. A. (ed.), *Fish Population Dynamics*. John Wiley, London: 83–103.
- Schneider D., Mustafić P., Mrakovčić M. & Mihaljević Z. 2000: Some aspects of the biology of the Neretvan spined loach. *Folia Zool.* 49 (Suppl.1): 159–165.
- Slavík O. & Ráb P. 1995: Effect of microhabitat on the age and growth of two stream-dwelling populations of spined loach, *Cobitis taenia*. *Folia Zool.* 44: 167–174.
- Taylor C.C. 1962: Growth equation with metabolic parameters. *J. Cons. Int. Expl. Mer.* 27: 270–286.
- Weatherley A.H. & Gill H.S. 1987: *The Biology of Fish Growth*. Academic Press, London.
- Zanella D., Mrakovčić M., Schneider D., Mustafić P., Čaleta M. & Radić I. 2003: Growth of *Cobitis narentana* Karaman, 1928 in the Neretva River, Croatia. *Folia Biol. (Kraków)* 51(Suppl.): 155–157.
- Zar J.H. 1984: *Biostatistical Analysis*. Englewood Cliffs, New Jersey, Prentice-Hall Inc.