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Short communication

Specific testicular morphology in the atherinomorph *Odontesthes nigricans* (Richardson, 1848): a comparison with the sympatric species *Odontesthes smitti* (Lahille, 1929)

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In teleosts, testis organization can be classified in different morphological categories (Grier, 1993; Grier and Parenti, 1994). While primitive osteichthyans show anastomosed tubular testes, the derived teleost species (*sensu* Parenti and Grier, 2004) present one of two principal morphologies according to the type of distribution of spermatogonia: spermatogonial non-restricted (SNR) or spermatogonial restricted (SR). The former is common in most teleosts, where the spermatogonia are distributed in the germinal epithelium along the seminiferous lobule length. The latter, with the spermatogonia restricted at the distal blind end of the testis lobule, is described only in Atheriniformes (Grier, 1981; Parenti and Grier, 2004).

The two atherinomorph species studied inhabit the South American marine coasts. *Odontesthes smitti* (Lahille, 1929) has been reported along the Atlantic Ocean from Uruguay (34°S) to Tierra del Fuego (54°S) and the Malvinas Islands (Cousseau and Perrotta, 2000), and along the Pacific Ocean coast from Tierra del Fuego to Seno Última Esperanza (52°S) (Dyer, 2000). *Odontesthes nigricans* (Richardson, 1848) is found on the Atlantic Ocean coast from Orense (39°S, Buenos Aires Province) south to Cabo de Hornos (56°S, Cape Horn) (Dyer, 2000).

The aims of the present study were to describe a specific structure observed in the testis of *O. nigricans* and to analyse the morphological differences with the testicular structure of the sympatric species *O. smitti*.

Odontesthes nigricans and *O. smitti* were caught together monthly on the Atlantic coast of Tierra del Fuego (Punta María: 54°S, 67°30′W) with trammel nets (50 m long, 2.5 m high, and 38 mm stretched mesh size). Transported on ice to the laboratory, the fishes were measured and dissected about 10 h after capture. Total length (T_L) and standard length (S_L) of each individual were measured with a digital calliper (±0.1 mm). Total body mass (M_T), gonad mass (M_G), liver mass (M_L) and gut mass (M_{GUT}) were recorded to the nearest 0.01 g. The gonadosomatic (eviscerated) index was calculated as $I_G = M_G\ 100\ M_T^{-1} - (M_G + M_L + M_{GUT})$.

For histological observations, 80 *O. smitti* and 131 *O. nigricans* testes were fixed in Bouin's solution, washed in water and transferred to 70% alcohol. Slides for microscopy were obtained by embedding transversal portions of approximately 5 mm of each testis in Paraplast®, sectioned (7–10 μm) and stained with PAS-haematoxylin. Seven specimens of each species with mature testes caught in November 2007 were used to determine the relationship between the area and perimeter of transversal testes sections. One histological section of each specimen was photographed with a Nikon Coolpix 5100 digital camera (Nikon Corp., Tokyo, Japan) and analysed using image analysis software (Image-Pro Plus®, version 3.0 Media Cybernetics). The perimeter (P) and area (A) of selected sections were measured to the nearest 10 μm and 100 μm^2, respectively, and the size corrected dividing by S_L.

Although *O. nigricans* and *O. smitti* testes are both elongated with a notorious blood vessel in the longitudinal axis, their macroscopic structures differ greatly. Whereas *O. smitti* has a smooth external surface (Fig. 1a), *O. nigricans* shows a deeply lobulated surface (Fig. 1b). The microscopic structure of mature testes of both species corresponds to the SR type. Histological cross-sections of *O. nigricans* testis showed the presence of a complex structure with furrows on its surface (Fig. 2b). These furrows delimit lobes with secondary collector ducts that connect radially with the main central spermatic ducts. Blind ends of lobules containing the spermatogonia are located near the external surface and attached beneath the tunica albuginea of the testis. The cysts containing spermatogonia and spermatocytes are close to the distal end of the seminiferous lobules. As spermatogenesis proceeds, the cysts with spermatids and spermatozoids are located progressively closer to primary collector ducts, which run centrally along the lobulations. Finally, spermatozoa are released into these collector ducts.

In contrast, histological cross-sections of *O. smitti* showed only a few striations on the testicular surface (Fig. 2a). Spermatogenesis in this species occurs as in *O. nigricans* within the seminiferous lobules, which in turn are arranged radially.

The I_G values for *O. nigricans* and *O. smitti* mature testes did not show any differences between species (t-test, $n = 14$, $P > 0.05$, Table 1). While the $A\ S_L^{-1}$ ratio of the testes cross-section did not differ between species (t-test, $n = 14$, $P > 0.05$), the $P\ S_L^{-1}$ ratio was higher in *O. nigricans* (t-test, $n = 14$, $P < 0.001$) due to the lobes shown by the testis of this species (Fig. 2b).

The external morphology and the microscopic structure of *O. smitti* are similar to other Atherinomorpha such as *Odontesthes (Basilichtys) bonariensis* (Valenciennes, 1835) (Calvo and Dadone, 1972), *Jenynsia lineata* (Jenyns, 1842) (Calvo and

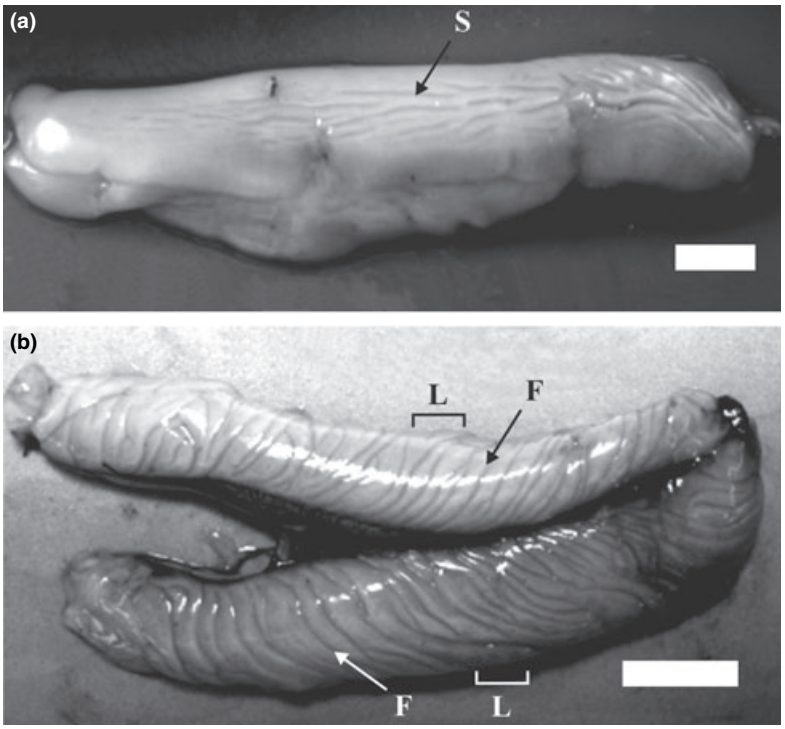

Fig. 1. A mature testis of (a) *Odontesthes smitti* and (b) *O. nigricans*. F, furrows; L, lobes; S, longitudinal striations. Bar = 1 cm

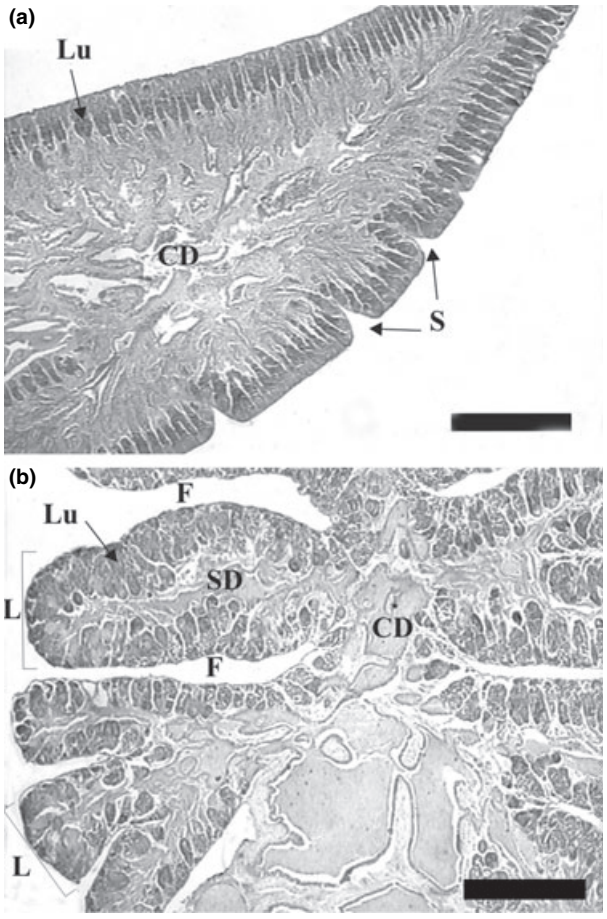

Fig. 2. Cross-section of a mature testis of (a) *Odontesthes smitti* and (b) *O. nigricans*. CD, central spermatic ducts; F, furrows; L, lobes; Lu, seminiferous lobules; S, longitudinal striations; SD, secondary ducts. Bar = 100 μm

Table 1
Sample size (N). Mean ± SD testicular area (A) and perimeter (P) corrected by standard length (S_L)

	N	$A\ S_L^{-1}$ ± SD	$P\ S_L^{-1}$ ± SD**	I_G ± SD
O. nigricans	7	0.25 ± 0.06	0.48 ± 0.14	11.27 ± 0.06
O. smitti	7	0.24 ± 0.05	0.21 ± 0.05	9.38 ± 2.22

Mean ± SD. Gonadosomatic Index (I_G) of *O. nigricans* and *O. smitti* (**$P < 0.001$).

Morriconi, 1972), *Jenynsia multidentata* (Jenyns, 1842) (Martinez and Monasterio Gonzo, 2002), *Austrolebias charrua* (Costa and Cheffe, 2001; Arezo et al., 2007), *Chirostoma jordani* Woolman, 1894 (Cardenas Reygadas and Barrera Escorcia, 1998) and *Chirostoma humboltianum* (Valenciennes, 1835) (Uria et al., 1998). In contrast, the *O. nigricans* testis surface with a great number of lobes was not previously described.

A good correlation between I_G values and sperm competition was found in several fishes species (Stockley et al., 1997; Taborsky, 1998; Stoltz and Neff, 2006). However, the use of I_G would be not appropriate to compare sperm production in *O. smitti* and *O. nigricans*. Although mature testis of both *Odontesthes* species showed similar I_G values, the amount of sperm produced in *O. nigricans* is assumed to be higher because of the larger perimeter of the histological cross-section of this species (Table 1). This morphology increases the space to lodge spermatogonia and, presumably, to augment the sperm production.

Although these two silverside species are the target of artisanal fisheries along the Patagonian coasts, the biological knowledge necessary to develop sustainable fisheries is still lacking. In this context, this is the first study on reproductive traits of these species in the southernmost extreme of their distribution. A more detailed approach is being conducted by

the authors, integrating the sex ratio and the spawning behaviour in *Odontesthes* populations, to improve comprehension of the functional value of the specific testis morphology as displayed by *O. nigricans*.

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