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Short Communication

Sympatric occurrence of two cytotypes of *Astyanax scabripinnis* (Characiformes, Characidae)

Edson Luis Maistro[1], Claudio Oliveira[2] and Fausto Foresti[2]

Abstract

Among four specimens of *Astyanax scabripinnis* sympatrically collected from the Tamanduá stream (State of São Paulo, Brazil), three, named cytotype I, had 2n = 50 chromosomes (6M + 26SM + 4ST + 14A) and a fundamental number (FN) of 86, and one specimen, named cytotype II, had 2n = 48 chromosomes (6M + 28SM + 4ST + 10A) and FN = 86. Besides the difference in diploid number, the cytotypes showed a clear difference in the distribution of constitutive heterochromatin. One NOR-bearing chromosome pair was detected in both cytotypes and one specimen of cytotype I had multiple NORs (tree chromosome pairs). Robertsonian translocations and an increase or loss of heterochromatin are proposed to explain the karyotypic divergence observed. Some aspects related to the chromosome evolution of *Astyanax scabripinnis* are discussed.

INTRODUCTION

The study of headwater fishes is interesting from an evolutionary viewpoint due to the occurrence of a small number of geographically isolated individuals in almost all species inhabiting these ecological niches. These characteristics facilitate the occurrence of speciation events (Britski, 1972; Moreira-Filho and Bertollo, 1991; Futuyma, 1993; Maistro *et al*., 1998).

Astyanax scabripinnis is a headwater species complex composed of local populations with several morphological (Fowler, 1948) and cytogenetic particularities (Moreira-Filho and Bertollo, 1991; Salvador and Moreira-Filho, 1992; Souza and Moreira-Filho, 1995; Souza *et al*., 1995; Mizoguchi and Martins-Santos, 1998; Maistro *et al*., 1998; among others). In the present investigation, we studied a sample of *A. scabripinnis*, which exhibited individuals with different diploid numbers and chromosome structures.

MATERIAL AND METHODS

A cytogenetic survey was performed on 4 specimens (3 females and 1 male) of *A. scabripinnis* collected from the Tamanduá stream, a small tributary of the Paranapanema River, located in the town of Itatinga, State of São Paulo, Brazil. The fishes were identified and are deposited in the fish collection of the Laboratório de Pesquisas em Genética, Universidade de Alfenas, Brazil. Mitotic cells were obtained from gill and kidney tissues by the air drying technique as described by Bertollo *et al.* (1978). Chromosome morphology was determined on the basis of arm ratios as proposed by Levan *et al.* (1964) and the chromosomes were classified as metacentrics (M), submetacentrics (SM), subtelocentrics (ST) and acrocentrics (A). The fundamental number (FN) was determined considering M, SM and ST chromosomes with two arms and A chromosomes with one arm. C-banding was performed by the method of Sumner (1972) and silver staining of nucleolus organizer regions (NORs) by the method of Howell and Black (1980).

RESULTS AND DISCUSSION

Cytogenetic studies have been carried out on several local populations of *A. scabripinnis* from different Brazilian regions. In these studies, three diploid numbers have already been detected: 2n = 46 for one population, 2n = 48 for five populations and 2n = 50 chromosomes for more than thirty populations (Moreira-Filho and Bertollo, 1991; Souza et al., 1995; Maistro et al., 1998, among others).

In the present study a total of 266 metaphases of four *A. scabripinnis* specimens were analyzed. Two different modal diploid numbers were detected, 2n = 50 (three specimens) and 2n = 48 chromosomes (one specimen), named cytotype I and cytotype II, respectively. Cytotype I had 3 pairs of metacentrics, 13 pairs of submetacentrics, 2 pairs of subtelocentrics and 7 pairs of acrocentrics, and FN = 86. Cytotype II had 3 pairs of metacentrics, 14 pairs of submetacentrics, 2 pairs of subtelocentrics and 5 pairs of acrocentrics, and FN = 86 (Figure 1).

The occurrence of a local population with more than one cytotype has been reported for several fish species

[1]*Instituto de Farmácia e Nutrição, Universidade de Alfenas, 37130-000 Alfenas, MG, Brasil. Send correspondence to E.L.M.*
[2]*Departamento de Morfologia, Instituto de Biociências, UNESP, Campus de Botucatu, 18618-000 Botucatu, SP, Brasil.*

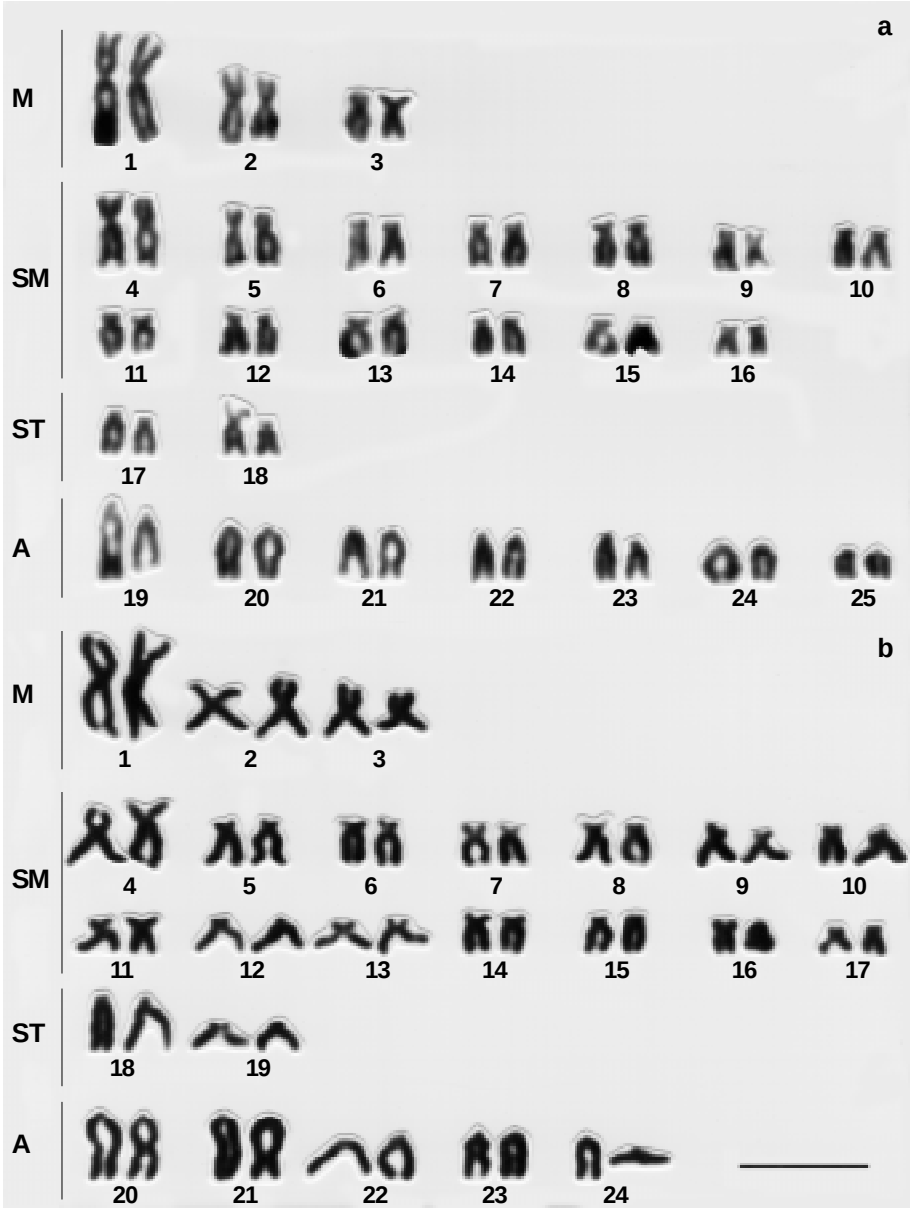

Figure 1 - Karyotypes of *A. scabripinnis* collected from the Tamanduá stream. **a**, Cytotype I (2n = 50), and **b**, cytotype II (2n = 48). Bar = 10 μm.

such as those of the genus *Bryconamericus* (Wasko *et al.*, 1996), in which different cytotypes had the same diploid number and different chromosome formulae. In other genera such as *Corydoras* (Oliveira *et al.*, 1990, 1993), *Gymnotus* (Fernandes-Matioli, 1996) and *Hoplias* (see a review in Bertollo *et al.*, 1997), cytotypes with different diploid numbers have been described. Among 37 populations of *A. scabripinnis* analyzed (Souza *et al.*, 1995; Maistro *et al.*, 1998), specimens with different diploid number were found in sympatry only in two, the population from Canta Galo brook (Souza *et al.*, 1995) and the population from Pardo River (Vieira, M.R., Oliveira, C. and Foresti, F., unpublished data).

The two cytotypes identified in the present study differed by the amount and distribution of constitutive heterochromatin. The karyotypes were composed of small heterochromatin blocks in the pericentromeric regions of most chromosomes of both cytotypes, and a conspicuous block in the terminal position on the long arm in one ST and three A pairs only in cytotype II. Pair number 19 (ST) exhibited a positive C-band segment on the short arm in cytotype II at the same position of the NOR region (Figure 2). This result agrees with the observation that the pattern of heterochromatin distribution has been the best discriminating feature among the local populations of *A. scabripinnis* (Moreira-Filho and Bertollo, 1991; Souza

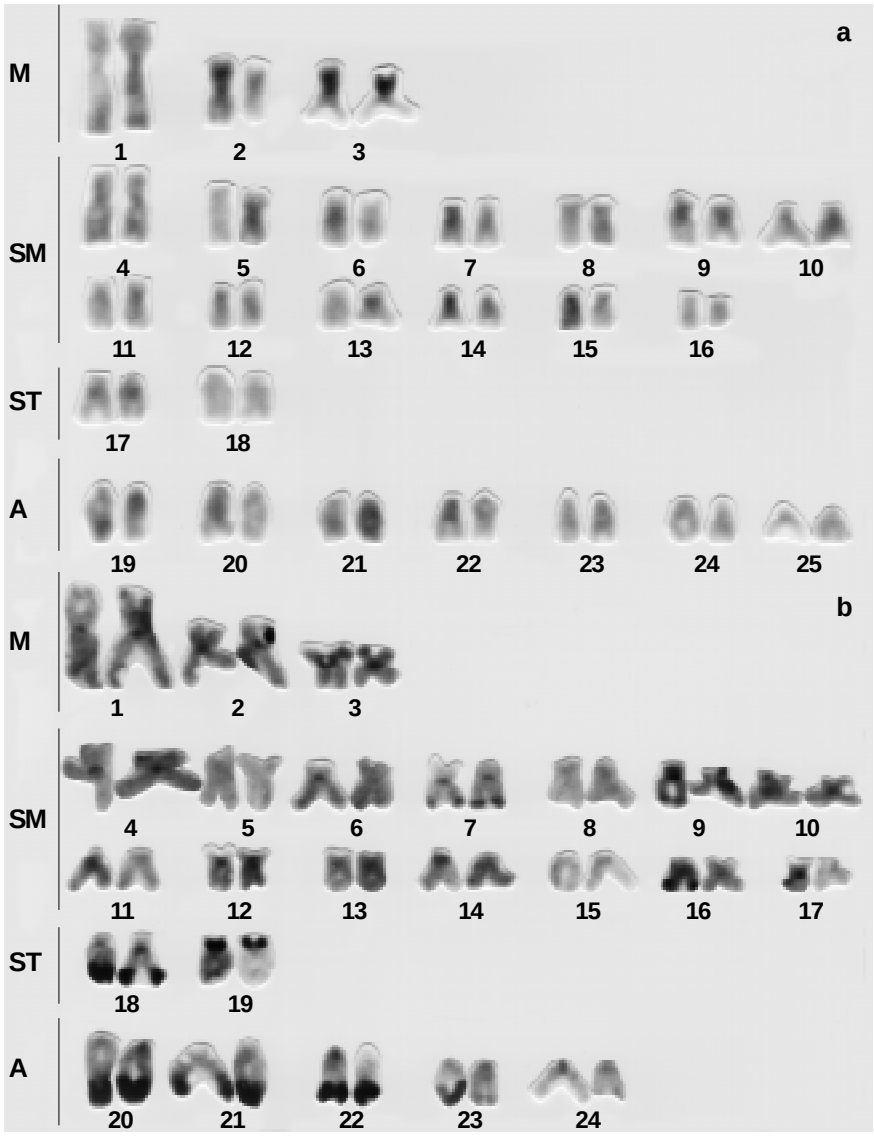

Figure 2 - C-banding karyotypes of *A. scabripinnis* from the Tamanduá stream. **a**, Cytotype I (2n = 50), and **b**, cytotype II (2n = 48).

and Moreira-Filho, 1995; Mizoguchi and Martins-Santos, 1998; Maistro *et al.*, 1998).

On both cytotypes one chromosome pair, a small-sized ST, had Ag-stained NORs in the terminal position on the short arm. However, one specimen of cytotype I showed two other chromosomes with Ag-NOR signals: an SM, with NORs in the terminal position on the long arm, and an M, with NORs in the terminal position on the short arm (Figure 3). The common NOR-bearing chromosome (ST) has been identified as the NOR-bearing one in several local populations of *A. scabripinnis* (Moreira-Filho, 1989; Souza and Moreira-Filho, 1995; Maistro et al., 1998), showing that this chromosome may be an important marker for this species. Besides this chromosome pair with NORs, several other chromosomes have been identified as NOR-bearing in *A. scabripinnis* (Maistro *et al.*, 1998). The largest number of NOR-bearing chromosomes in *A. scabripinnis* was described for a sample from the Jucu River which had 13 NOR-bearing chromosome pairs (Rocon-Stange and Almeida-Toledo, 1993).

Considering the chromosome formulae of the two cytotypes identified in the present study and the fact that they probably had a common ancestor, the best hypothesis to explain this divergence is the occurrence of a Robertsonian translocation between two acrocentric chromosome pairs of a fish with 50 chromosomes, followed by an increase or loss of heterochromatin. An alternative hypothesis is that the cytotypes found in the Tamanduá River originated from different ancestral stocks that only recently began to live in sympatry. A similar hypothesis has

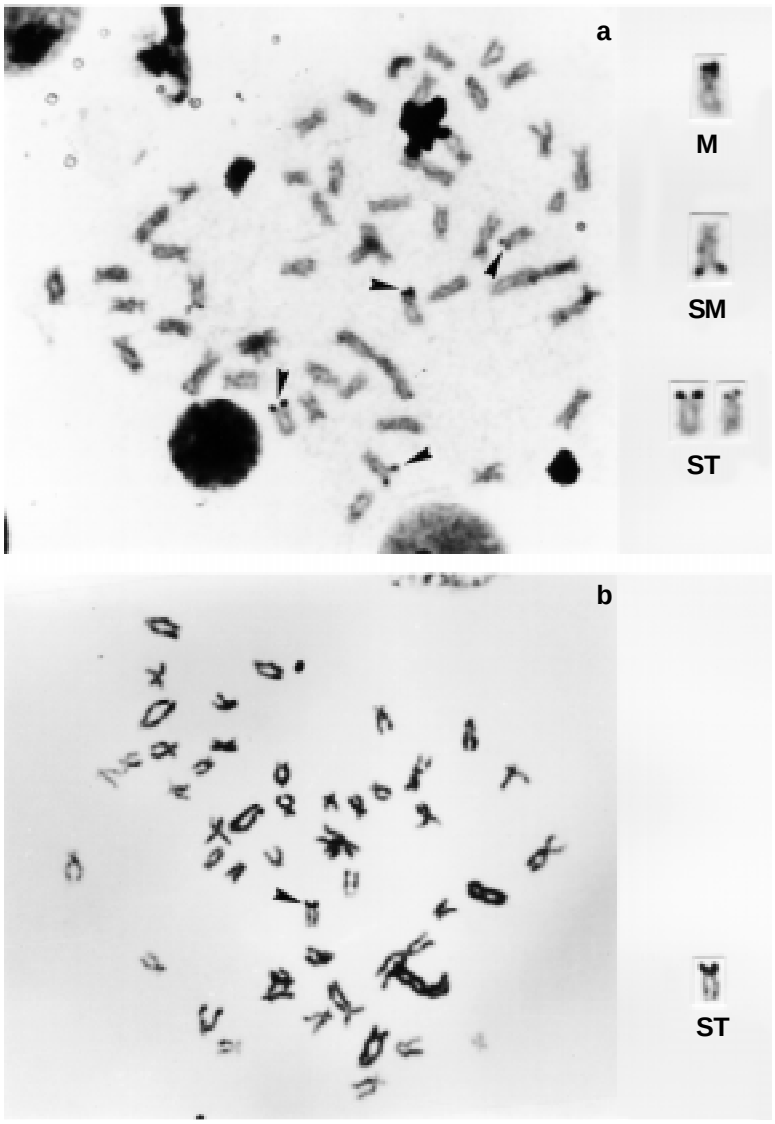

Figure 3 - Somatic metaphases of *A. scabripinnis* from the Tamanduá stream showing the NOR-bearing chromosomes (arrowheads). **a**, Cytotype I (2n = 50) and **b**, cytotype II (2n = 48).

been proposed to explain the existence of two cytotypes of *A. scabripinnis* living in sympatry in the Canta Galo brook (Souza *et al*., 1995) but new data will be necessary to confirm it.

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RESUMO

Foram analisados citogeneticamente quatro exemplares de *Astyanax scabripinnis* coletados simpatricamente no ribeirão Tamanduá (Estado de São Paulo, Brasil). Três deles, denominados de citótipo I, apresentaram 2n = 50 cromossomos (6M + 26SM + 4ST + 14A) e um número fundamental (FN) igual a 86, e um exemplar, denominado de citótipo II, apresentou 2n = 48 cromossomos (6M + 28SM + 4ST + 10A) e FN = 86. Ao lado das diferenças no número diploide, os citótipos mostraram uma clara diferença na distribuição da heterocromatina constitutiva. Um par de cromossomos portadores de RONs foi detectado nos dois citótipos e um exemplar com o citótipo I apresentou RONs múltiplas (três pares cromossômicos). Translocações Robertsonianas e aumento ou perda de heterocromatina são propostas para explicar a divergência cariotípica observada. Alguns aspectos relacionados à evolução cromossômica em *Astyanax scabripinnis* são discutidos.

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