



Cytogenetic analysis of three catfish species of the family Pseudopimelodidae (Teleostei, Siluriformes)

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Abstract

Cytogenetic analyses performed in *Cephalosilurus apurensis*, *Microglanis* aff. *cottoides* and *Pseudopimelodus bufonius* revealed that the three species have $2n = 54$ chromosomes: *C. apurensis* presented six metacentric (M), 28 submetacentric (SM), 14 subtelo-centric (ST), and six acrocentric (A) chromosomes, while *M. aff. cottoides* showed 10M, 32SM, 10ST and 2A, and *P. bufonius* had 12M, 30SM and 12ST. The nucleolus organizer regions (NORs) were present on the short arm of a middle-sized ST pair, identified as pair 19, in *C. apurensis* NORs were found on the short arm of a middle-sized ST (pair 23) and on the long arm of a middle-sized ST (pair 22) in *M. aff. cottoides* and on the short arm of three middle-sized ST pairs, identified as pairs 9, 10 and 11, in *P. bufonius*. C-banding revealed a very small amount of constitutive heterochromatin in the chromosomes of all species, including the NORs. The occurrence of $2n = 54$ in all species of the family Pseudopimelodidae and its absence among species of the closely related Pimelodidae and Heptapteridae may be important in identifying Pseudopimelodidae species.

Key words: karyotype, chromosomes, C-banding, Ag-NOR, fish.

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The order Siluriformes (catfishes) has 3,093 species, divided into 36 families and 478 genera, and distributed worldwide, except for the coldest areas in the Northern and Southern hemispheres (Ferraris, 2007). Recent phylogenetic studies showed that the old family Pimelodidae comprised three monophyletic units: Pimelodidae, Heptapteridae and Pseudopimelodidae (Lundberg *et al.*, 1991; de Pinna, 1998). According to Ferraris (2007), Pseudopimelodidae is composed of the genera *Batrochoglanis* (five species), *Cephalosilurus* (four species), *Lophiosilurus* (one species), *Microglanis* (14 species) and *Pseudopimelodus* (five species). A new genus and species, *Cruciglanis pacifisi*, has been recently described by Ortega-Lara and Lehmann (2006). This family is widely distributed in South America and is considered the least known family among the naked Neotropical freshwater catfishes (Shibatta, 2003). Currently, the only species to have their karyotypes reported are *Microglanis garavello* (cited as *M. cottoides* - Vissotto *et al.*, 1999a) and *Pseudopimelodus mangurus* (Martinez *et al.*, 2004). The objective of the present study

was to analyze the karyotypes of *Cephalosilurus apurensis*, *Microglanis* aff. *cottoides* and *Pseudopimelodus bufonius*.

The following specimens were karyotyped: one male specimen of *Cephalosilurus apurensis* from the Orinoco River, Caicara del Orinoco, Bolívar, Venezuela (07°38'11.6" N, 66°19'04.2" W, LBP 3034); two males and four females of *Microglanis* aff. *cottoides* from Ribeirão Cavalto Stream, Jaraguá do Sul, Santa Catarina, Brazil (26°28,250' S, 49°10,958' W, LBP 731) and two males and two females of *Pseudopimelodus bufonius* from the Amazon (aquarium trade, LBP 2345). The specimens were identified and deposited in the fish collection of the Laboratório de Biologia e Genética de Peixes (LBP), Departamento de Morfologia, Instituto de Biociências, Universidade Estadual Paulista, São Paulo, Brazil.

Mitotic chromosome preparations were obtained according to the technique described by Foresti *et al.* (1993). Nucleolar organizer regions (Ag-NORs) were revealed by the silver-staining method (Howell and Black, 1980) and C-banding was performed according to Sumner (1972). The chromosomes were classified according to their arm ratios as metacentrics (M), submetacentrics (SM), subtelo-centrics (ST), and acrocentrics (A) (Levan *et al.*, 1964).

The three species analyzed possessed $2n = 54$ chromosomes. *Cephalosilurus apurensis* had 6M, 28SM, 14ST and 6A (Figure 1a), *Microglanis* aff. *cottoides* presented 10M, 32SM, 10ST and 2A (Figure 2a) and *Pseudopimelodus bufonius* showed 12M, 30SM and 12ST (Figure 3a). A $2n = 54$ is characteristic for the family Pseudopimelodidae and the karyotypes of *C. apurensis* and *M. aff. cottoides* are similar to those observed in other species of the family, which typically have chromosomes of all morphological types, except for *M. garavelloi* and *P. bufonius* that do not have any acrocentric chromosome (Table 1).

The $2n = 54$ present in Pseudopimelodidae contrasts with the modal $2n = 56$ found in most catfish families (Oliveira and Gosztanyi, 2000) and specially with the diploid numbers found among representatives of Heptapteridae and Pimelodidae, which are closely related to Pseudopimelodidae (Sullivan *et al.*, 2006) (Table 1).

Pseudopimelodid species have single or multiple Ag-NORs (Table 1). *Cephalosilurus apurensis* showed a single pair of Ag-NORs on the short arms of a middle-sized ST pair, identified as pair 19 (Figure 1a). The remaining pseudopimelodid species analyzed also had a single Ag-NOR: *Lophiosilurus alexandrii* showed Ag-NORs on

the short arm of a SM (Marques, Garcia and Moreira Filho, personal communication); *Microglanis garavelloi* (Vissotto *et al.*, 1999a) had Ag-NORs on the long arm of M; and *Pseudopimelodus mangurus* (Martinez *et al.*, 2004) presented Ag-NORs on the short arm of SM/ST (Table 1). Single Ag-NORs were also identified in all species of Pimelodidae and all but one species of Heptapteridae (Table 1). This is also the most common condition in Siluriformes (Oliveira and Gosztanyi, 2000) and even in Teleostei (Klinkhardt, 1998). The Ag-NORs of *M. aff. cottoides* were found on the short arm of a middle-sized ST pair,

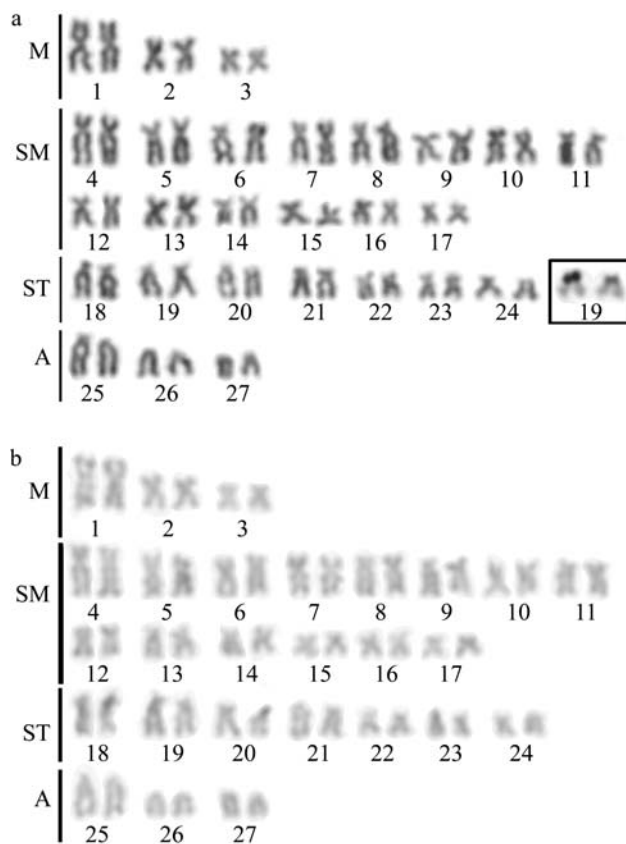


Figure 1 - Karyotype of *Cephalosilurus apurensis* ($2n = 54$) after: (a) conventional staining and (b) C-banding. In the inset, silver stained chromosomes showing the terminal Ag-NOR on the short arms of pair 19.

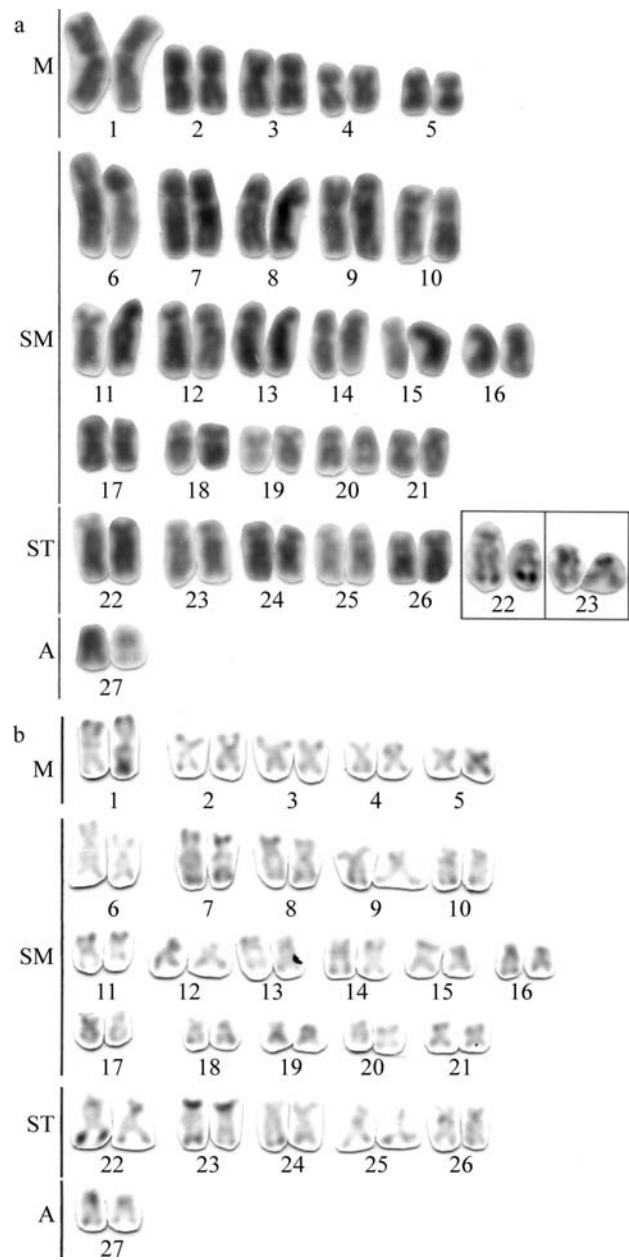


Figure 2 - Karyotype of *Microglanis* aff. *cottoides* ($2n = 54$) after: (a) conventional staining and (b) C-banding. In the inset, silver stained chromosomes showing the terminal Ag-NORs on the short arms of pair 23 and long arms of pair 22.

Table 1 - Cytogenetic data for Pseudopimelodidae, Heptapteridae and Pimelodidae.

Family/species	Locality	2n	Karyotype	NOR	References
Pseudopimelodidae					
<i>Cephalosilurus apurensis</i>	Orinoco River, Caicara del Orinoco, Bolívar, Venezuela	54	6M+28SM+14ST+6A	1	Present study
<i>Lophiosilurus alexandrii</i>	Três Marias Reservoir, Minas Gerais, Brazil	54	54M, SM, ST, A	1	Marques, Garcia and Moreira Filho (personal communication)
<i>Microglanis aff. cottoides</i>	Ribeirão Cavalto Stream, Jaraguá do Sul, Santa Catarina, Brazil	54	10M+32SM+10ST+2A	2	Present study
<i>Microglanis garavelloii</i>	Araquá and Capivara Rivers, Botucatu, São Paulo, Brazil	54	22M+20SM+12ST	1	Vissotto <i>et al.</i> (1999a)
<i>Pseudopimelodus bufonius</i>	Amazon Basin	54	12M+30SM+12ST	3	Present study
<i>Pseudopimelodus mangurus</i>	Mogi-Guaçu River, Pirassununga, São Paulo, Brazil	54	6M+26SM+12ST+10A	1	Martinez <i>et al.</i> (2004)
Heptapteridae					
<i>Pimelodella avanhandavae</i>	Araquá River, São Paulo, Brazil	46	20M+20SM+6ST	1	Vissotto <i>et al.</i> (1999a)
<i>Pimelodella aff. meeki</i>	Couro do Boi River, Paraná, Brazil	46	34M+12ST	1	Dias and Giuliano-Caetano (2002)
<i>Heptapterus longicauda</i>	Quinta Stream, Itatinga, São Paulo, Brazil	52	22M+26SM+4ST	2	Vissotto <i>et al.</i> (1999a)
<i>Pimelodella aff. avanhandavae</i>	Tibagi River, Paraná, Brazil	52	30M+22SM	1	Swarça <i>et al.</i> (2003a)
<i>Imparfinis cf. piperatus</i>	Juquiá River, Juquiá, São Paulo, Brazil	56	22M+26SM+4ST+4A	1	Vissotto <i>et al.</i> (2001)
<i>Rhamdella microcephala</i>	Machado River, São João da Mata, Minas Gerais, Brazil	56	18M+30SM+8ST, A	1	Fonseca <i>et al.</i> (2003)
<i>Cetopsorhamdia iheringi</i>	Capivara River, Botucatu, São Paulo, Brazil	58	28M+24SM+6ST	1	Vissotto <i>et al.</i> (1999a)
<i>Imparfinis mirini</i>	Quinta Stream, São Paulo, Brazil	58	M24M+34SM/F23M+35SM	1	Vissotto <i>et al.</i> (1997)
<i>Imparfinis piperatus</i>	Araras River, Araras, São Paulo, Brazil	58	32M+26SM	1	Vissotto <i>et al.</i> (2001)
<i>Pimelodella kronei</i>	Iporanga, São Paulo, Brazil	58	54M, SM+4ST	1	Almeida-Toledo <i>et al.</i> (1992)
<i>Pimelodella transitoria</i>	Iporanga, São Paulo, Brazil	58	54M, SM+4ST	1	Almeida-Toledo <i>et al.</i> (1992)
<i>Rhamdia quelen</i>	Quadros Lagoon, Rio Grande do Sul, Brazil	58	52M, SM, ST+6A	1	Hochberg and Erdtmann (1988)
Pimelodidae					
<i>Calophysus macropterus</i>	Negro River, Amazonas, Brazil	50	22M+18SM+10A	1	Ramirez-Gil <i>et al.</i> (1998)
<i>Pirinampus pinirampu</i>	Tibagi River, Sertaneja, Paraná, Brazil	50	26M+12SM+2ST+10A	1	Swarça <i>et al.</i> (1999)
<i>Pseudoplatystoma fasciatum</i>	Solimões River, Amazonas, Brazil	56	18M+14SM+10ST+14A	1	Fenocchio and Bertollo (1992)
<i>Pseudoplatystoma tigrinum</i>	Solimões River, Amazonas, Brazil	56	18M+16SM+8ST+14A	1	Fenocchio and Bertollo (1992)
<i>Sorubim lima</i>	Solimões River, Amazonas, Brazil	56	18M+12S+14ST+12A	1	Fenocchio and Bertollo (1992)
<i>Bergtaria westermanni</i>	São Francisco River, Minas Gerais, Brazil	56	42M, SM+14ST	1	Dias and Foresti (1993)
<i>Pimelodus heraldoi</i>	Tibagi River, Paraná, Brazil	56	22M+22SM+6ST+6A	1	Souza <i>et al.</i> (2004)
<i>Pimelodus maculatus</i>	São Francisco River, Minas Gerais, Brazil	56	40M, SM+16ST, A	1	Dias and Foresti (1993)
<i>Pimelodus argenteus</i>	Paraguai River, Corumbá, Mato Grosso do Sul, Brazil	56	34M, SM+22ST, A	1	Souza <i>et al.</i> (2003)
<i>Pimelodus misteriosus</i>	Paraguai River, Corumbá, Mato Grosso do Sul, Brazil	56	26m+20SM+2ST+8A	1	Souza <i>et al.</i> (2003)
<i>Pseudoplatystoma corruscans</i>	Porto Rico, Paraná, Brazil	56	18M+16SM+10ST+12A	1	Martins-Santos <i>et al.</i> (1996)
<i>Hemisorubim platyrhynchos</i>	Porto Rico, Paraná, Brazil	56	22M+18SM+6ST+10A	1	Martins-Santos <i>et al.</i> (1996)
<i>Zungaro zungaro</i>	Foz do Iguaçu, Paraná, Brazil	56	26M+10SM+6ST+14A	1	Martins-Santos <i>et al.</i> (1996)
<i>Iheringichthys labrosus</i>	Jurumirim Reservoir, Itatinga, São Paulo, Brazil	56	22M+18SM+10ST+6A	1	Vissotto <i>et al.</i> (1999b)
<i>Steindachneridion sp.</i>	Iguaçu River, Usina Salto Segredo, Paraná, Brazil	56	20M+24SM+2ST+10A	1	Swarça <i>et al.</i> (2003b)

2n = diploid number; M = metacentrics; SM = submetacentrics; ST = subtelocentrics; A = acrocentrics; NOR = number of chromosome pairs with nucleolar organizer regions.

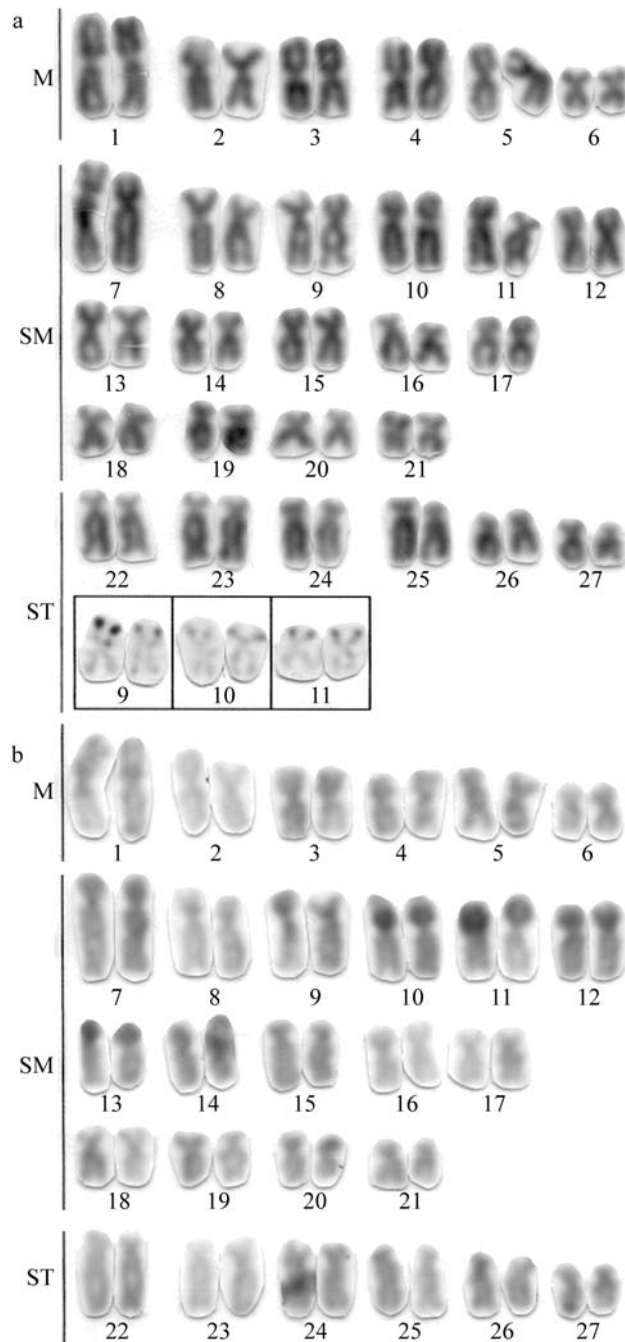


Figure 3 - Karyotype of *Pseudopimelodus bufonius* ($2n = 54$) after: (a) conventional staining and (b) C-banding. In the inset, silver stained chromosomes showing the terminal Ag-NORs on the long arms of pairs 9, 10 and 11.

identified as pair 23, and on the long arm of another middle-sized ST pair, identified as pair 22 (Figure 2a). The Ag-NORs of *P. bufonius* occurred on the short arm of three middle-sized ST pairs, identified as pairs 9, 10 and 11 (Figure 3a). Multiple Ag-NORs were identified in one species of Heptapteridae (Vissotto *et al.*, 1999a) and were not found among Pimelodidae (Table 1). The number and position of NORs are species-specific and do not seem to follow any pattern during karyotypic evolution.

C-banding showed the occurrence of a small amount of constitutive heterochromatin in the chromosomes of the three species (Figures 1b, 2b, 3b). In *Cephalosilurus apurensis*, positive C-banded segments were observed on the short arms of the largest ST pair (pair 19) and in the Ag-NORs. In *Pseudopimelodus bufonius*, C-banding evidenced segments on the short arms of the six larger SM pairs (pairs 9, 10, 11, 12, 13 and 14) and in *Microglanis aff. cottoides*, C-banding revealed positive segments on the short arms of one large ST pair (pair 23) and on the long arms of several ST pairs. The small amount of heterochromatic segments in the chromosomes of *P. bufonius*, *M. aff. cottoides*, and *C. apurensis*, as well as in other representatives of the family Pseudopimelodidae, *P. mangurus* (Martinez *et al.*, 2004), and *M. garavelloii* (Vissotto *et al.*, 1999a), suggests that this may be a characteristic of this catfish family. The occurrence of a very small amount of C-banded positive segments reported herein resembles the data reported for many teleost species, including siluriforms (Gold *et al.*, 1990).

The presence of $2n = 54$ chromosomes may be an important characteristic to differentiate Pseudopimelodidae species from species of Heptapteridae and Pimelodidae. Further analysis of additional Pseudopimelodidae species with different staining techniques will provide important information for a better understanding of the chromosome evolution in the group and will help to confirm the conservative nature of the diploid number in this fish family.

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