

CYTOGENETIC STUDIES ON FISHES OF THE FAMILY PIMELODIDAE (SILUROIDEI)

Ana Lúcia Dias and Fausto Foresti

ABSTRACT

Four fish species of the family Pimelodidae were analyzed. *Bergiaria westermanni* and two different *Pimelodus* species have the same diploid chromosome number ($2n = 56$). Despite some differences in chromosome structure, these species are highly similar in karyotype and differ from *Pimelodella* sp., which presents a reduction in chromosome number to $2n = 46$. The data confirm the extensive chromosome variability existing in this family, characterized by intraindividual and/or population polymorphisms of a structural nature which may or may not be sexlinked, and by the presence of supernumerary chromosomes.

INTRODUCTION

Among fishes of the suborder Siluroidei, approximately 100 species belonging to 12 families have been investigated for chromosome number (Legrande, 1982; Oliveira *et al.*, 1988). Most of the representatives of these siluroid families present diploid numbers of $2n = 56 \pm 2$, $FN = 80$ and a frequently asymmetrical karyotype, including elements with one and two arms. However, each family was found to have a broad range of chromosome numbers per species, so that it has been difficult to establish a general pattern of chromosome diversification in relation to the phyletic lineage of siluroids (Kim *et al.*, 1982).

The first cytogenetic studies on the family Pimelodidae were conducted by Toledo and Ferrari (1976a,b) who analyzed three species belonging to the genus *Pimelodus*, one belonging to the genus *Pimelodella* and one belonging to the genus

Rhamdia. Further chromosome analyses have been recently performed on Pimelodidae (Hochberg and Erdtmann, 1983; Hochberg *et al.*, 1985; Almeida Toledo *et al.*, 1985; Mestriner and Galetti Jr., 1986; Hochberg and Erdtmann, 1988; Fenocchio and Bertollo, 1990, 1992), revealing a diploid number ranging from 54 to 58 in the species studied.

We analyzed the karyotypes and chromosome characteristics of four Pimelodidae species.

MATERIAL AND METHODS

Chromosome characteristics of the following fish species of the Pimelodidae family were analysed: *Pimelodus maculatus*, 10 males and 10 females collected from the Mogi-Guaçu River and six males and nine females collected from the São Francisco River; *Pimelodella* sp., nine males and 10 females from the Mogi-Guaçu River; *Pimelodus* sp., eight males and nine females, and *Bergiaria westermanni*, four males and three females, both from the São Francisco River.

Mitotic preparations were obtained from kidney cells, especially from the anterior portion of the organ, using the air drying technique described by Foresti *et al.* (1981). The nucleolar organizer regions were identified by the silver staining technique of Howell and Black (1980) and the constitutive heterochromatin segments were visualized by the C banding technique of Sumner (1972).

The karyotypes were arranged in chromosome pairs in decreasing order of size and grouped into meta-submetacentric (M-SM) and subtelo-acrocentric (ST-A) sets according to the criteria of Levan *et al.* (1964).

RESULTS

Genus Pimelodus

The *P. maculatus* specimens from the Mogi-Guaçu River and the *P. maculatus* and *Pimelodus* sp. specimens from the São Francisco River presented a modal diploid value equal to 56 chromosomes, with the karyotype characterized by 20 chromosome pairs of the M-SM group and eight pairs of the ST-A group, for a total number of chromosome arms (NF) equal to 96. No differences in chromosome complement were observed between sexes (Figures 1 and 2).

However a small difference in chromosome structure was observed in a male specimen of *P. maculatus*, i.e., a small metacentric, not present in the standard karyotype, was observed in all cells analyzed, apparently substituting a large chromosome of the ST-A group (Figure 3A). In *Pimelodus* sp., two specimens, a male and a female, presented a similar situation (Figure 3B). In this case, a large chromosome of the ST-A group was

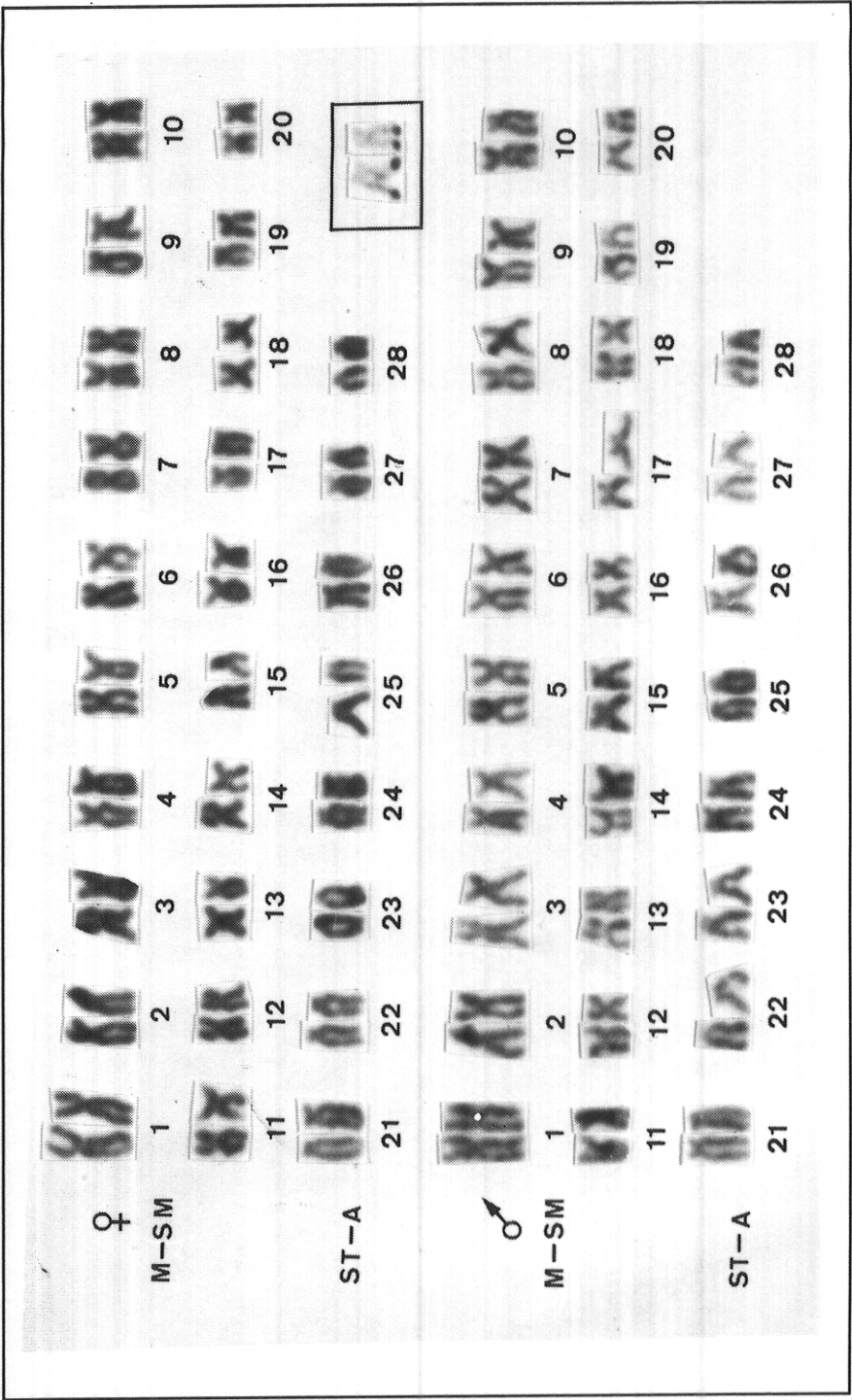
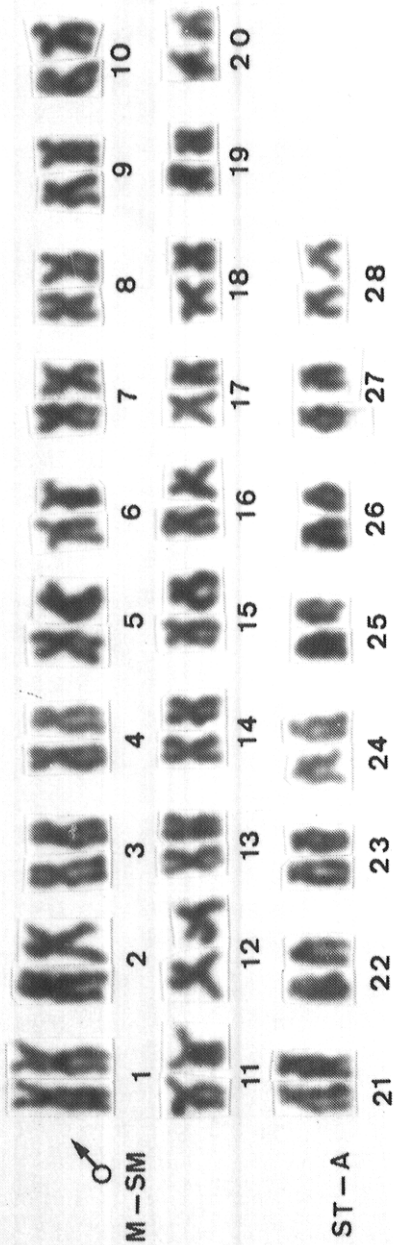
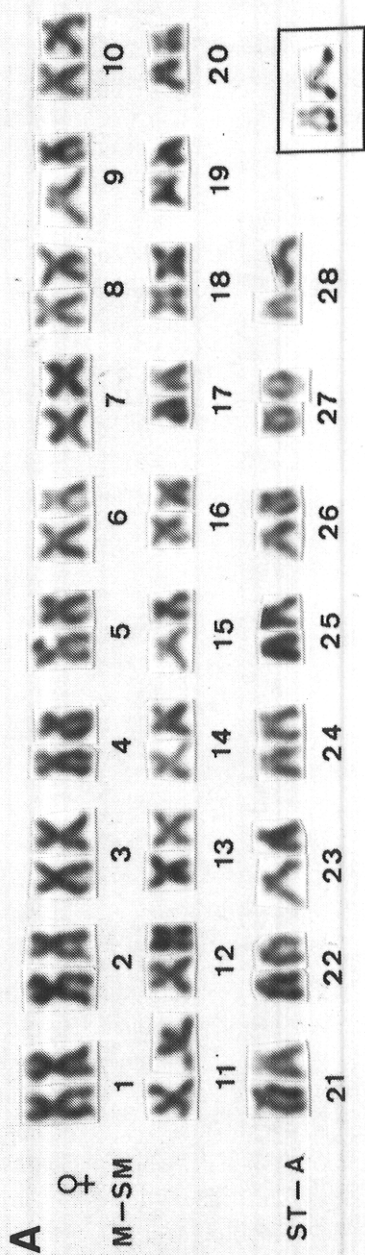


Figure 1 - Karyotypes of females and males of *Pimelodus maculatus* from the Mogi-Guaçu River. In the inset, the Ag-NOR-bearing pair (possibly pair 27).



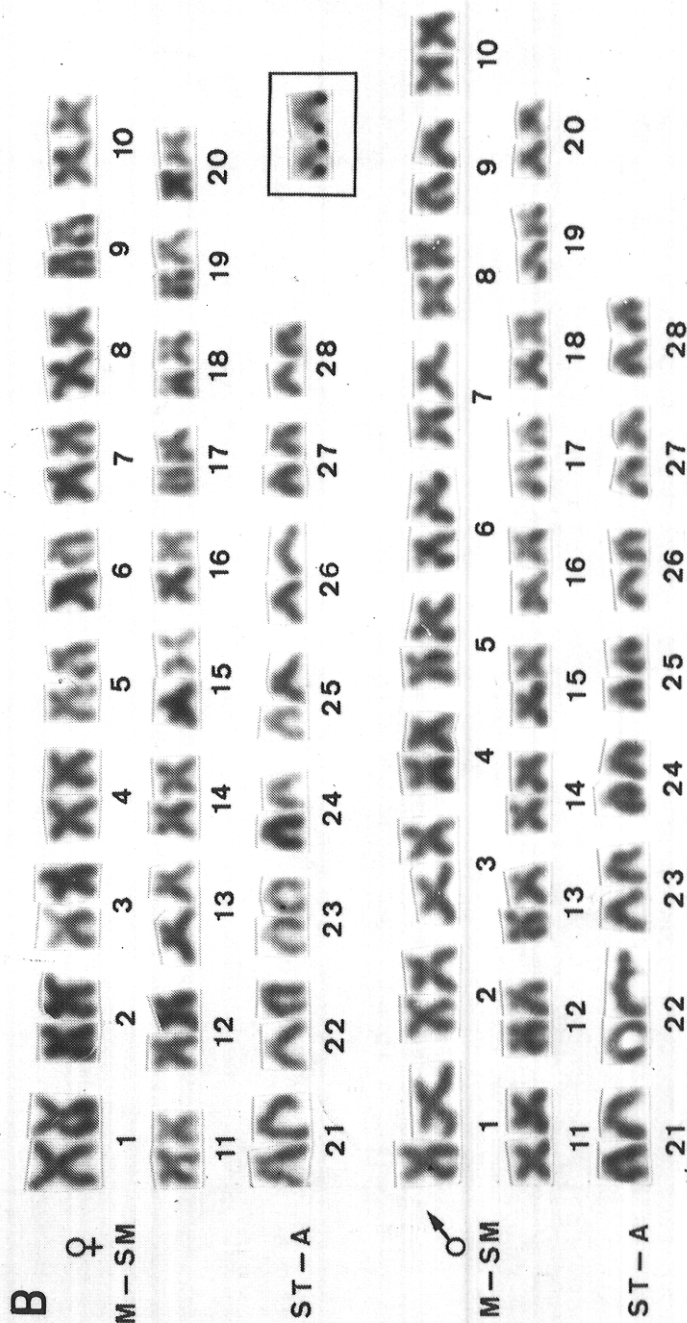


Figure 2 - Karyotypes of females and males of *Pimelodus maculatus* (A) and *Pimelodus* sp. (B) from the São Francisco River. In the insets, the Ag-NOR-bearing pairs (possibly pair 27) in both species.

A



M-SM

1	2	3	4	5	6	7	8	9	10
XX	XY	XX	XX	XX	XX	XX	XX	XX	XX

11	12	13	14	15	16	17	18	19	20
XX	XX	XX	XX	XX	XX	XX	XX	XX	XX

ST-A

21	22	23	24	25	26	27	28
XX	XX	XX	XX	XX	XX	XX	XX

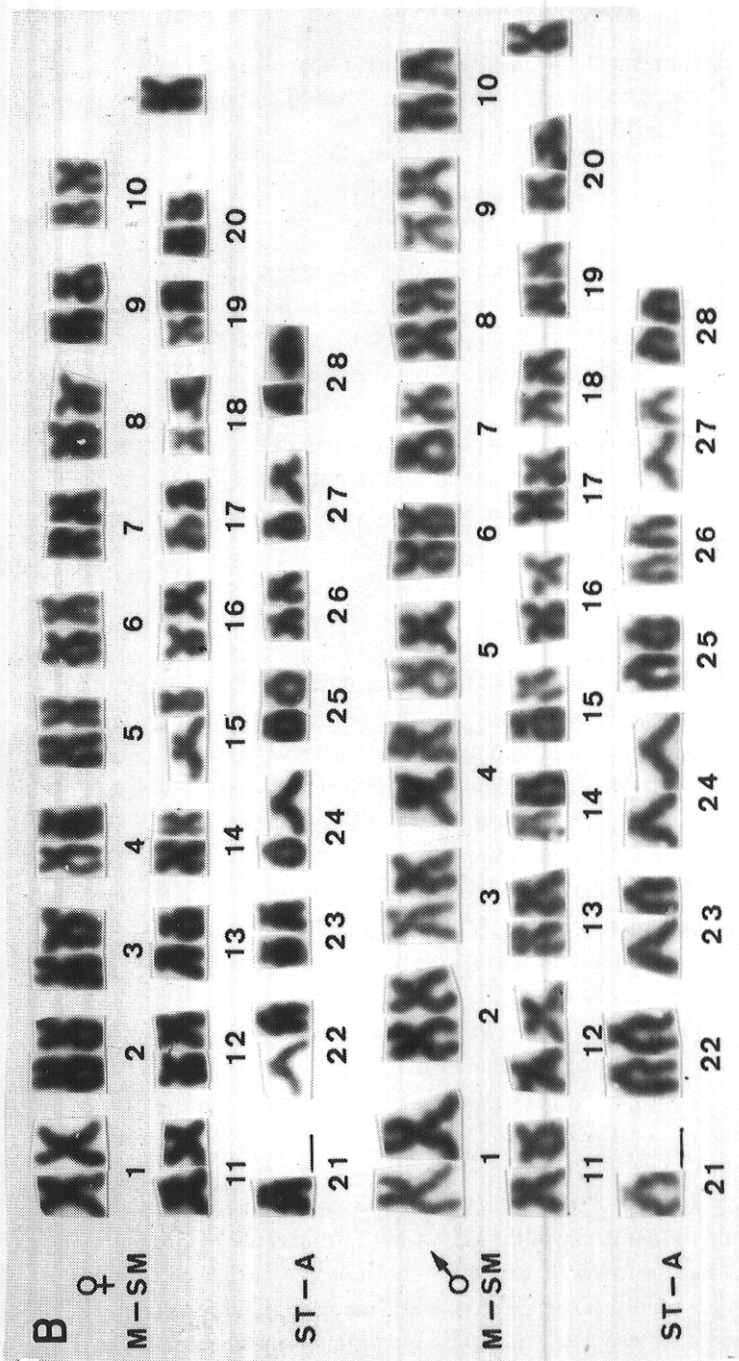


Figure 3 - Karyotypes of *Pimelodus maculatus* (male) from the São Francisco River (A) with 20 M-SM pairs + 1 M and 7 ST-A pairs + 1 ST; and *Pimelodus* sp. (female and male) from the São Francisco River (B) showing the same karyotypic formulae. Two types of unpaired chromosomes are observed in the karyotypes of both species, independent of sex.

apparently replaced by a medium-sized metacentric. In these species, the nucleolar organizer region (NOR) was located on the end of the longer arm of a chromosome pair of the ST-A group, possibly pair no. 27 (Figures 1 and 2).

Genus Pimelodella

In *Pimelodella* sp. from the Mogi Guaçu River, specimens of both sexes presented the same diploid number of $2n = 46$. The female karyotype consisted of 20 M-SM pairs and 3 ST-A pairs, whereas the male karyotype consisted of 19 M-SM pairs plus a large submetacentric chromosome and another small metacentric in addition to three ST-A pairs (Figure 4).

This species has a medium-sized nucleolar chromosome pair of the M-SM group and presents extensive intra- and interindividual variability with respect to NOR size and staining intensity. Two or three adjacent silver-stained regions were observed on the same chromosome segment in some cells (Figure 5).

Genus Bergiaria

The *B. westermanni* specimens from the São Francisco River presented a diploid number of $2n = 56$ for both sexes, with a karyotype consisting of 21 M-SM chromosome pairs and seven ST-A pairs (Figure 6). In all the specimens, the occurrence of small chromosomes was observed in somatic cells from males and females (Figure 7), with the characteristics of B or supernumerary chromosomes, which were found to be fully heterochromatic when submitted to C banding (Figure 8).

Joint analysis of the data for male and female specimens of *B. westermanni* showed that cells with two B were more frequent, followed by cells with three and four B. The number of cells with the minimum (zero) and maximum (five) number of supernumerary chromosomes was very low.

The NOR of this species was located on the terminal portion of the larger arm in an ST-A chromosome, probably pair no. 27 (Figure 6).

DISCUSSION

Analysis of the karyotypes of *P. maculatus* and *Pimelodus* sp. demonstrated a diploid number of $2n = 56$ and the similarity of the karyotypic formulas (20 M-SM pairs and 8 ST-A pairs) showed that no marked structural chromosome variations occur between these species. It is possible that the medium-sized metacentric chromosomes exclusively found in the variant karyotypes, which presented a small metacentric in the place of a large chromosome of the group ST-A, originated from a pericentric inversion

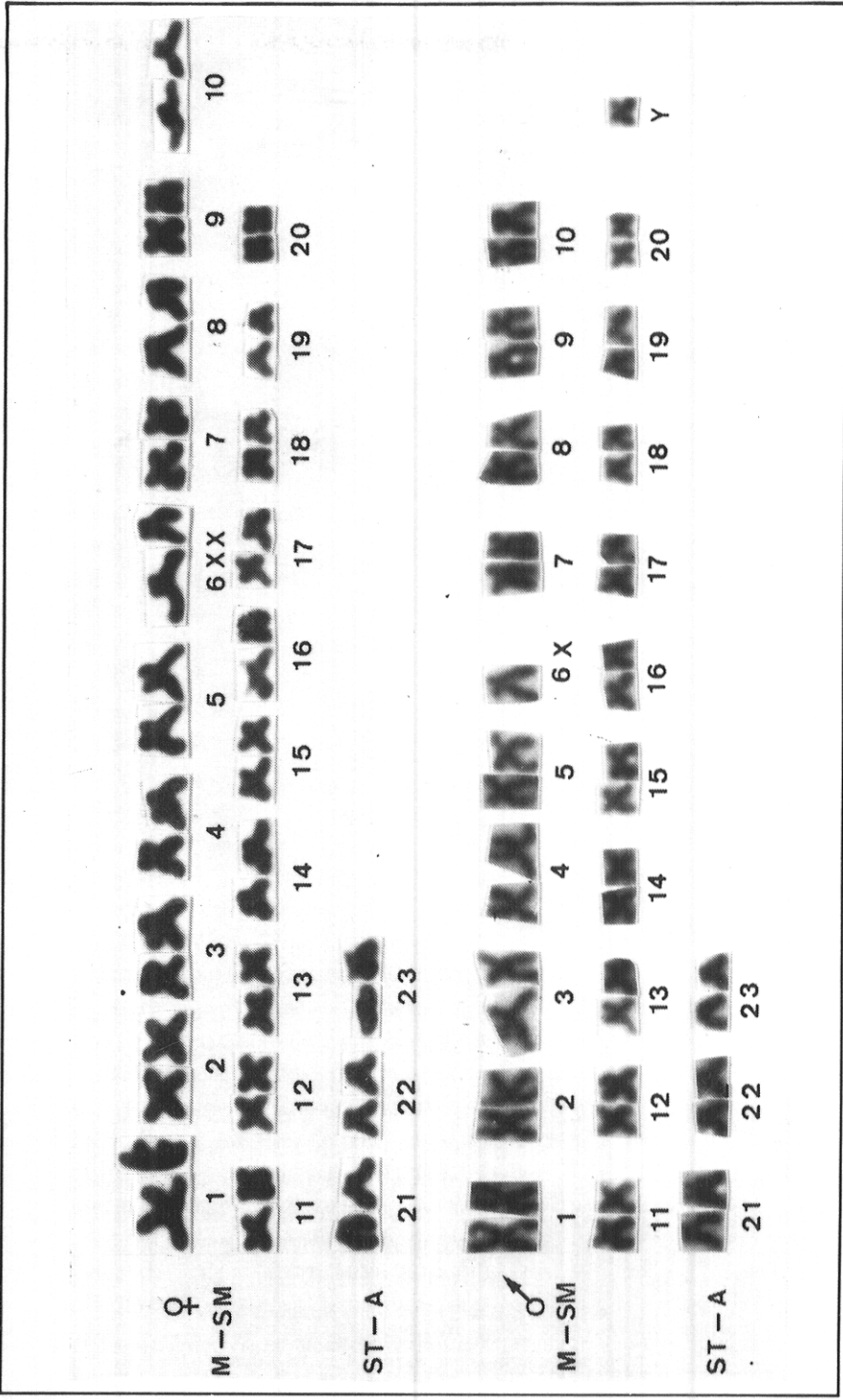


Figure 4 - Karyotypes of females and males of *Pimelodella* sp. from the Mogi-Guaçu River. Male karyotypes show a heteromorphic chromosome pair.

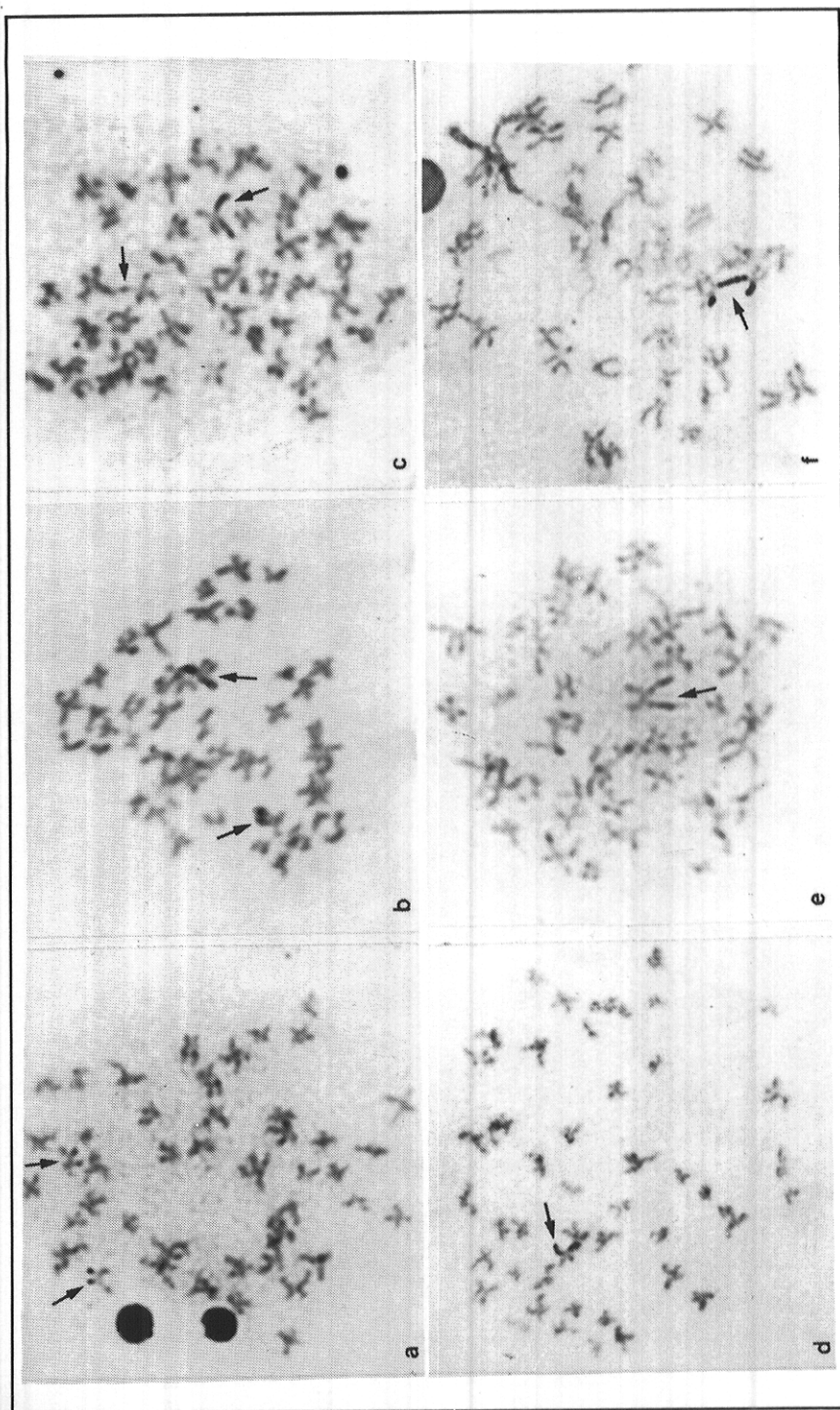


Figure 5 - Ag-NORs in *Pimelodella* sp. Only one chromosome pair is involved in nucleolar organization in this species, which presents extensive polymorphism. Metaphase cells (a-f) show different patterns of size and number of NOR segments. The NORs are indicated by arrows.

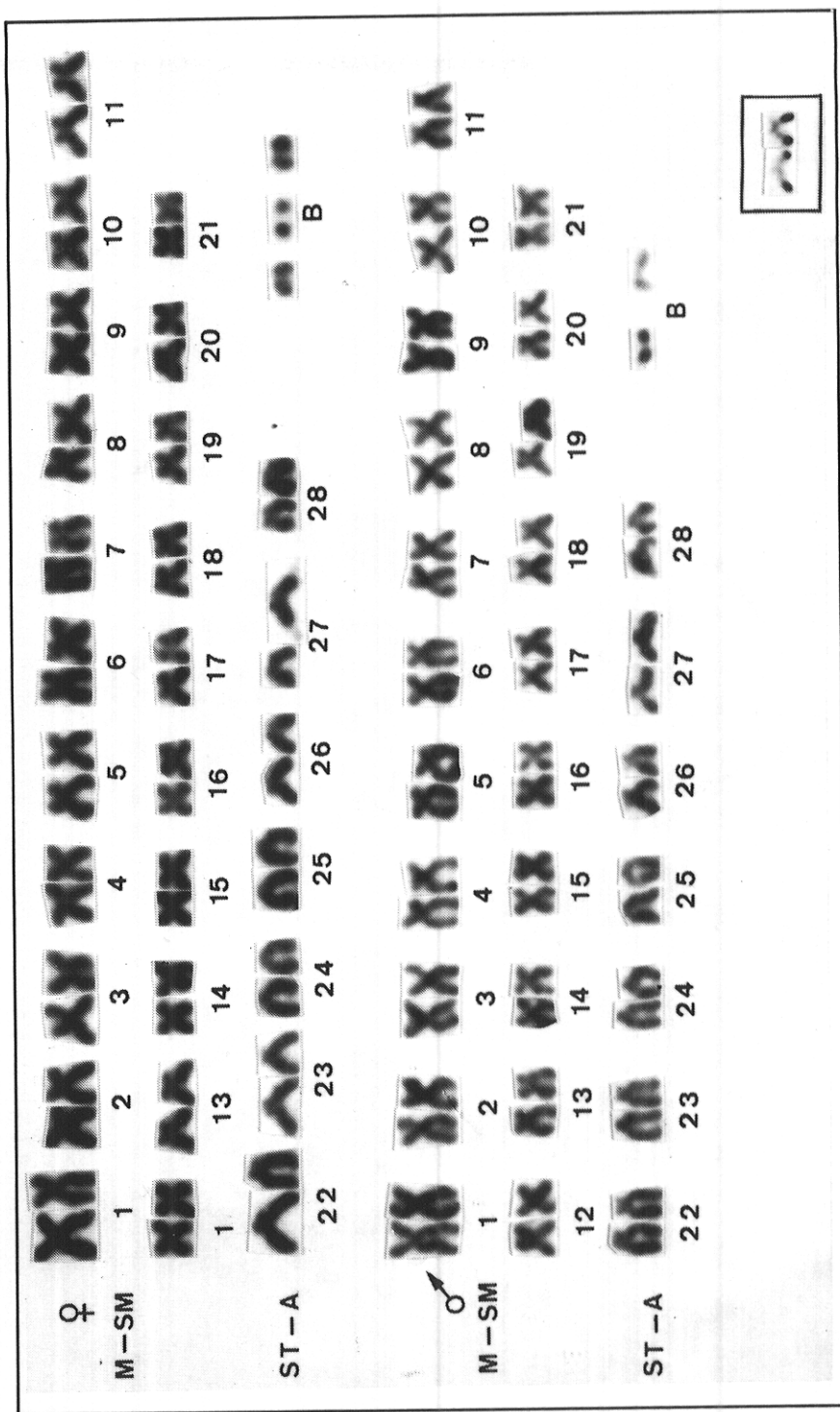


Figure 6 - Karyotypes of females and males of *Bergiaria westermanni* from the São Francisco River presenting supernumerary chromosomes (B). In the inset, the Ag-NOR-bearing pair (possibly pair 27).

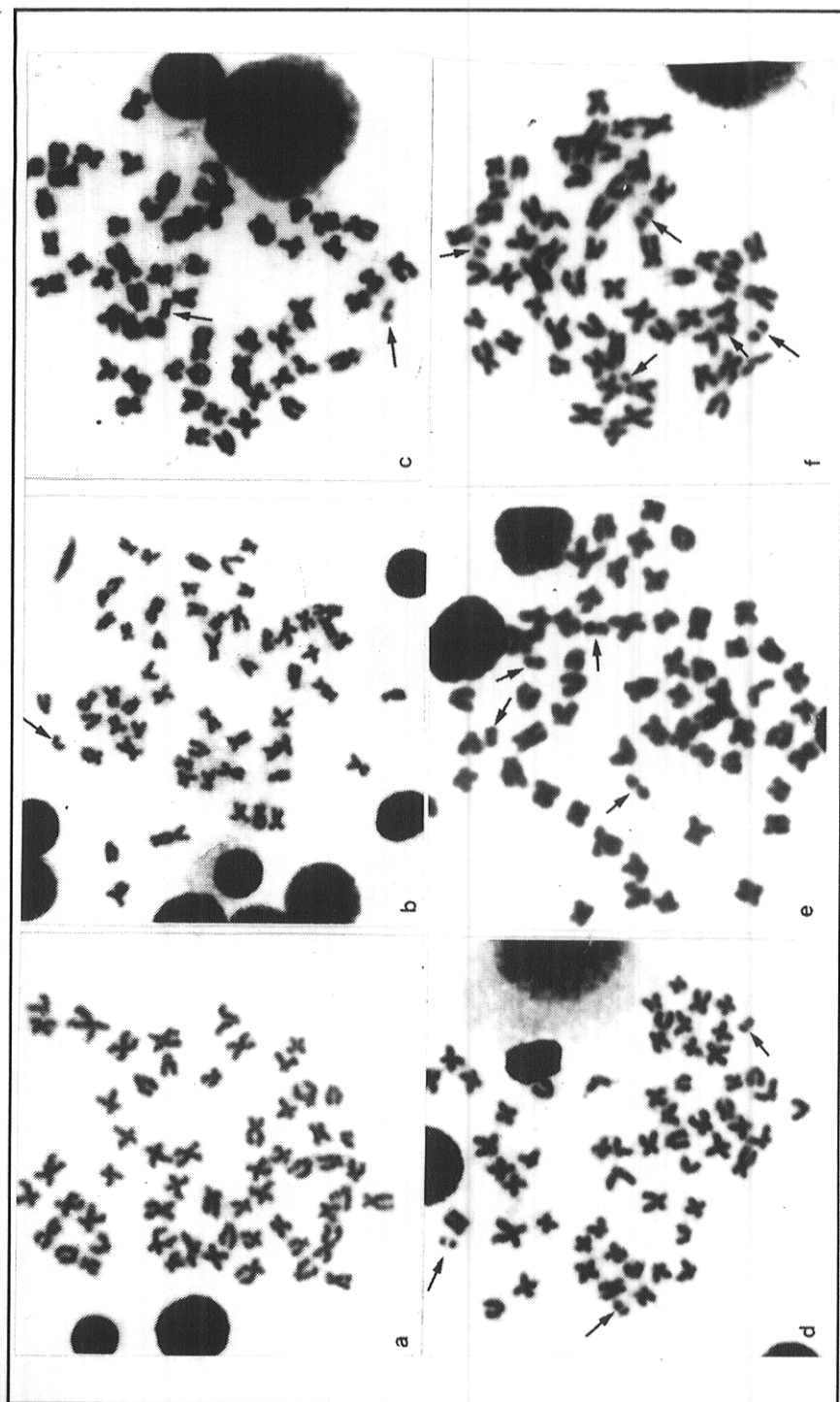


Figure 7 - Somatic metaphase cells in different individuals of *Bergiaria westermanni* from the São Francisco River. Different numbers of supernumerary chromosomes were found (0 to 5) and are indicated by arrows.

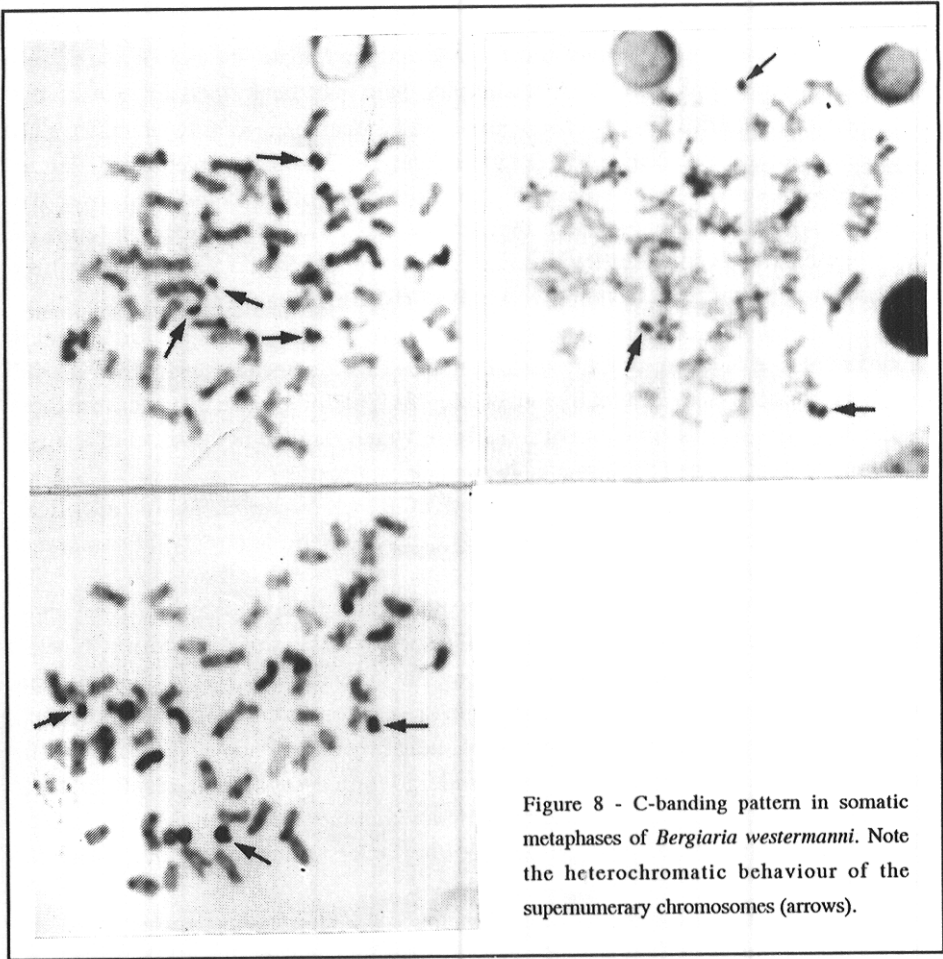


Figure 8 - C-banding pattern in somatic metaphases of *Bergiaria westermanni*. Note the heterochromatic behaviour of the supernumerary chromosomes (arrows).

in an chromosome of the group ST-A. The reduction in size of the new metacentric chromosome may then have occurred by subsequent small deletions.

Our data extend the limits of variation of chromosome number in Pimelodidae. These results are quite similar to those observed in other families of the Siluroidei group. LeGrande (1981) considers the chromosome evolution of this group to occur in parallel in different families, with a reduction of the diploid value from an ancestral form with $2n = 56 \pm 2$ chromosomes. If the same criterion of interpretation is used to explain the chromosome changes of Pimelodidae, the karyotype of *Pimelodella* sp. would be derived in relation to those of *Pimelodus* and *Bergiaria*. Furthermore, it is probable that

Pimelodella has a sex chromosome system of the XX/XY type, with the male representing the heterogametic sex with 44 autosomes + XY and the female, the homogametic sex with 44 autosomes + XX. This heteromorphic pair may be formed by one of the chromosomes in pair 6, a large submetacentric, and a small metacentric, which have no corresponding chromosomes in the female karyotype. However, Toledo and Ferrari (1976b) did not detect this heteromorphism in specimens of *Pimelodella* sp. from the same environment, probably because they used a less efficient technique in terms of quality of the chromosome preparations. The number of M-SM and ST-A chromosomes we found was also different from that reported by these investigators.

In the family Pimelodidae, *Pimelodella* sp. appears to be the first species showing evidence of heteromorphic sex chromosomes with mechanisms of the XX/XY type. It is possible that differentiation of the sex pair originated from a structural modification with the loss of chromatin material, which resulted in the onset of a small metacentric chromosome (Y) from a large submetacentric of pair 6.

The small chromosomes observed in *B. westermanni*, characterized as supernumerary, are similar to those detected by Pauls and Bertollo (1990) in *Prochilodus scrofa* and *P. cearensis* and also observed in other species of fish (Foresti *et al.*, 1989; Salvador and Moreira Filho, 1992). In the metaphases analyzed by the C-banding technique, these chromosomes unequivocally appear to be small chromatin blocks, revealing their heterochromatic nature. Among the individuals analyzed, there was an inter- and intraindividual numerical variation in these chromosomes. The intraindividual variability observed in the occurrence of supernumerary chromosomes (0B to 5Bs/cell) suggests that these chromosomes may undergo processes which result in their maintenance and possibly in their accumulation within the population. Mechanisms of mitotic and meiotic non-disjunction may be the factors determining this variability (Jones, 1975).

It appears that the modal diploid value of $2n = 56$ is the basic value of the Pimelodidae karyotype (Oliveira *et al.*, 1988; Fenocchio and Bertollo, 1992). Extensive chromosome variability occurs in the species of this group, characterized by intraindividual and/or population polymorphisms of a structural nature (sex linked or not), by the presence of supernumerary chromosomes and even by an effective reduction of the chromosome complement to $2n = 46$ in *Pimelodella*.

ACKNOWLEDGMENTS

This research was supported by CNPq, CAPES and the Federal University of São Carlos (UFSCar). The authors are grateful to CQDEVASF and CEPTA for providing the specimens for analysis.

Publication supported by FAPESP.

RESUMO

Foram analisadas quatro espécies de peixes da família Pimelodidae. *Bergiaria westermanni* e duas diferentes espécies de *Pimelodus* apresentam o mesmo número cromossômico ($2n = 56$). Apesar da existência de algumas diferenças na estrutura cromossômica, os cariótipos destas espécies são altamente similares e diferem do de *Pimelodella* sp., que apresenta uma redução do número cromossômico para $2n = 46$. Os dados confirmam a extensiva variabilidade cromossômica existente nesta família, caracterizada por polimorfismos dos tipos intraindividual e/ou intrapopulacional de natureza estrutural, ligados ou não ao sexo e pela presença de cromossomos supernumerários.

REFERENCES

- Almeida Toledo, L.F., Trajano, E., Toledo Filho, S.A. and Foresti, F. (1985). Análise citogenética comparativa entre o peixe cego, *Pimelodella kronei* das grutas de Iporanga (SP) e o seu presumível ancestral *Pimelodella transitoria*. *Ciênc. Cult.* 37: 777.
- Fenocchio, A.S. and Bertollo, L.A.C. (1990). Supernumerary chromosome in a *Rhamdia hilarii* population (Pisces, Pimelodidae). *Genética* 81: 193-198.
- Fenocchio, A.S. and Bertollo, L.A.C. (1992). Karyotype similarities among Pimelodidae (Pisces, Siluriformes) from the Brazilian Amazon region. *Cytobios* 69: 41-46.
- Foresti, F., Almeida Toledo, L.F. and Toledo, S.A. (1981). Polymorphic nature of nucleolus organizer regions in fishes. *Cytogenet. Cell Genet.* 31: 137-144.
- Foresti, F., Almeida Toledo, L.F. and Toledo, S.A. (1989). Supernumerary chromosome system, C-banding pattern characterization and multiple nucleolus organizer regions in *Moenkhausia sanctaefilomenae* (Pisces, Characidae). *Genética* 79: 107-114.
- Hochberg, V.B.M. and Erdtmann, B. (1983). Investigações citotaxonômicas em espécies do gênero *Rhamdia* (Pisces, Pimelodidae). Os jundiás da lagoa dos Quadros, RS. *Ciênc. Cult.* 35: 671.
- Hochberg, V.B.M. and Erdtmann, B. (1988). Cytogenetical and morphological considerations on *Rhamdia quelen* (Pisces, Pimelodidae). The occurrence of B chromosomes and polymorphic NOR regions. *Rev. Brasil. Genet.* 11: 563-576.
- Hochberg, V.B.M., Erdtmann, B. and Leistner, S. (1985). Caracterização de cromossomos supranumerários ou B em jundiás (Pisces, Pimelodidae). *Ciênc. Cult.* 37: 733.
- Howell, W.M. and Black, D.A. (1980). Controlled silver-staining of nucleolus organizer regions with a protective colloidal developer: a 1-step method. *Experientia* 36: 1014-1015.
- Jones, R.N. (1975). B chromosome systems in flowering plants and animal species. *Int. Rev. Cytol.* 40: 1-100.
- Kim, D.S., Park, E. and Kim, J.S. (1982). Karyotypes of nine species of the Korean catfishes. *Korean J. Genetics* 4: 57-68.
- LeGrande, W.H. (1981). Chromosomal evolution in North American catfishes (Siluriformes: Ictaluridae) with particular emphasis on the madtoms. *Noturus*. *Copeia* 1: 33-52.

- Levan, A., Fredga, K. and Sandberg, A.A. (1964). Nomenclature for centromeric position on chromosomes. *Hereditas* 52: 201-220.
- Mestriner, C.A. and Galetti Jr., P.M. (1986). Observações preliminares sobre os cromossomos de *Lophiosilurus alexandri* e suas relações com o grupo Pimelodidae. In: *Simpósio de Citogenética Evolutiva e Aplicada de Peixes Neotropicais*. São Carlos, Resumos; 49.
- Oliveira, C., Almeida Toledo, L.F., Foresti, F., Britski, H. and Toledo Filho, S. (1988). Chromosome formulae of neotropical freshwater fishes. *Rev. Brasil. Genet.* 11: 577-624.
- Pauls, E. and Bertollo, L.A.C. (1990). Distribution of a supernumerary chromosome system and aspects of cariotypic evolution in the genus *Prochilodus* (Pisces, Prochilodontidae). *Genetica* 81: 117-123.
- Salvador, L.B. and Moreira-Filho, O. (1992). B chromosomes in *Astyanax scabripinnis* (Pisces, Characidae). *Heredity* 69: 50-56.
- Sumner, A.T. (1972). A simple technique for demonstrating centromeric heterochromatin. *Exp. Cell Res.* 75: 304-306.
- Toledo, V. and Ferrari, I. (1976a). Estudo citogenético de 3 espécies do gênero *Pimelodus* (Pimelodinae, Pisces). *Cientifica* 4: 101-106.
- Toledo, V. and Ferrari, I. (1976b). Estudo citogenético de *Pimelodella* sp e *Rhamdia hilarii* (Pimelodinae, Pimelodidae, Pisces): cromossomo marcador. *Cientifica* 4: 120-123.

(Received November 3, 1992)