A Heteromorphic ZZ/ZW Sex Chromosome System in Fish, Genus Hypostomus (Loricariidae)

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Summary Ten specimens of Hypostomus sp. from the Araguaia basin (Barra do Garcas, MT, Brazil) were cytogenetically studied, with characterization of the constitutive heterochromatin and positive segments for the fluorochrome Mithramycin. It was detected a chromosomal heteromorphism related to sex, of the ZZ/ZW type, not observed yet in Hypostomus catfish. Thus, in male karyotype can be found one pair of subtelo-acrocentric Z chromosomes, represented by a unique component in female karyotype. However, this latter has an exclusive metacentric—the W chromosome. The available data indicate that the differentiation of this sex chromosome system may have occurred by a pericentric inversion, followed by length reduction in an ancestral chromosome similar to the Z one, originating a W chromosome like that now observed in the female karyotype of this fish species.

Key words Catfish, Hypostomus, Sex chromosomes

The Loricariidae, commonly known as catfish, are the second largest neotropical fish family in number of species (Britski 1972). Hypostomus is one of the most specious genus in this family, with more than 130 species already described and having a broad distribution (Isbrucker 1980). This group has been characterized as not conservative from a chromosomal viewpoint, concerning its diploid number, karyotypic macrostructure and chromosome banding, suggesting that different rearrangements have took place in the karyotype diversification and, consequently, in the evolutionary history of this genus (Artoni and Bertollo 1996).

Few Loricariidae species present sex chromosomes cytologically differentiated. Particularly in the subfamily Hypoptopomatinae there are species with male heterogamety (XX/XY), as Pseudotocinclus tietensis, and others with female heterogamety (ZZ/ZW), as Microlepdogaster leucofrenatus, suggesting an independent evolution concerning these two sex chromosome systems (Andreata et al. 1992, 1993). Loricariichthys platymetopon, represents another Loricariidae species with a ZZ/ZW sex chromosome system, different from that showed by Microlepdogaster leucofrenatus. Thus, while in this last case, the W chromosome differentiation probably occurred through a heterochromatin accumulation process, in Loricariichthys platymetopon it seems to have originated from a pericentric inversion (Scavone and Julio Jr. 1995).

So, among Loricariidae, it was observed at least three occurrences of heteromorphic sex chromosomes, with distinct origin and differentiation. On the other hand, among Anostomidae fish, several Leporinus species can be easily recognized by a ZZ/ZW sex chromosome system (Galetti and

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Foresti 1986), characterizing a monophyletic group inside this family (Galetti et al. 1995). Thus, it is possible that similar indications can be also obtained for other fish groups, as new cases of sex chromosomes are described, with characterization of evolutionary lines.

In the present study it is described the karyotype of *Hypostomus* sp. from the Araguaia river (MT-Brazil), showing a ZZ/ZW sex chromosome system that appears to be the first occurrence of such a type among the *Hypostomus* fish.

Materials and methods

Ten specimens of *Hypostomus* sp. (7 males and 3 females), from the Fundo stream, a small affluent of the Araguaia river (Barra do Garças, Mato Grosso state, Brazil) were studied. This is a species with problematical identification, as several others from this fish group, which may have its specific status better defined with the aid of karyotypic data.

The chromosomal preparations were obtained by the “air drying” standard method, using kidney cells (Bertollo et al. 1978). The nucleolus organizer regions were evidenced by silver nitrate (Howell and Black 1980) and the constitutive heterochromatin by the C-band technique (Sumner 1972). Mithramycin A was used for detecting fluorescent GC-rich regions, according Schmid (1980). Chromosomes were organized in three groups in the karyotype (metacentric: M, submetacentric: SM and subtelo/acrocentric: ST/A) and arranged in decreasing order of size, according the previous criteria proposed for this fish group (Artoni and Bertollo 1996).

Results and discussion

*Hypostomus* sp. from the Araguaia basin showed 2n=64 chromosomes in all specimens analyzed. The male karyotype presents 14M+24SM+26ST/A, while the female one shows 15M+24SM+25ST/A chromosomes (Fig. 1). Thus, a comparative analysis among male and female kary-
otypes evidences a sex-associated heteromorphism, the female representing the heterogametic sex in a ZZ/ZW sex chromosome system. The Z chromosome belongs to the subtelo/acrocentric group, while the W is a metacentric one. However the precise identification these two chromosomes inside their respective groups is not possible in standard stained preparations. Thus, their position in the karyotype (Fig. 1) is only a possible location. Although Hypostomus fish can be characterized by a non-conservative karyotype evolution (Artoni and Bertollo 1996), no sex chromosome system had still been identified in this genus. A possible XX/XY system was proposed by Michelle et al. (1977) for a species of Plecostomus (=Hypostomus), which requires a reanalysis and confirmation.

The nucleolus organizer regions are located on the short arm of a small ST/A pair (no. 30), which can show a conspicuous secondary constriction (Fig. 1). These regions were evidenced both by silver nitrate (not showed), and Mithramycin A (Fig. 2C, D). Thus, the proposition that NORs correspond to GC-rich regions in lower vertebrates (Schmid and Guttenbach 1988), is also observed in this Hypostomus species. Small blocks of centromeric heterochromatin occur in most of the chro-

Fig. 2. A and B: C-banded female and male metaphases, showing one and two Z chromosomes respectively (arrows and details). Note the prominent interstitial C-band on the long arm. The W chromosome does not present a particular C-banding pattern. C and D: Mithramycin stained metaphases showing the fluorescent positive NORs (arrows), in a heteromorphic and homomorphic condition, respectively.
mosome pairs, as evidenced by C-banding, besides some other telomeric bands (Fig. 2A, B). Differently from the other chromosomes of the karyotype, the Z chromosome present an interstitial C+ block located on its long arm (Fig. 2A, B). Thus, this specific chromosome mark makes possible its identification among the other chromosomes of the complement. Two chromosomes with this characteristic band occur in male specimens, while in females only one of such chromosome is observed. On the other hand, the W chromosome was not differentiated by the chromosomal banding methods here employed, so its identification is only based on morphological characteristics.

At present, two propositions try to explain the differentiation of sex chromosomes. One of them implies, at first, in the occurrence of a structural rearrangement, decreasing the recombination rate among the original homologous chromosomes (Beçak et al. 1969, among others). This rearrangement can be followed by a heterochromatinization process, with an increase or even loss of chromosomal material. In the other case, the heterochromatinization may represent the first step in the sex chromosome differentiation, related with the origin and multiplication of a satellite DNA (Ray-Chaudhuri et al. 1971, Singh et al. 1980, among others). The latter has been considered a suitable hypothesis to explain the differentiation of some fish sex chromosome systems, as in Leporinus (Galetti and Foresti. 1986), Triportheus (Bertollo and Cavallaro 1992), Parodon (Moreira-Filho et al. 1993) and Hypoptopomatinae (Andreata et al. 1992, 1993). Structural rearrangements, on the other hand, seem to be related to multiple sex chromosome systems, as in Apareiodon (Moreira-Filho et al. 1980, 1993), Eigenmannia (Almeida-Toledo et al. 1984) and Hoplias (Bertollo et al. 1997). In Hypostomus sp. a pericentric inversion in an original acrocentric chromosome similar to the Z one, followed by loss of chromatin material, would be a reasonable hypothesis to explain the differentiation of the W chromosome. These events would lead to the actual metacentric W chromosome, smaller than the Z, and without the interstitial constitutive heterochromatin (Figs. 1, 2). Thus, like in Loricairichthys platymetopon (Scavone and Julio Jr. 1995), a structural rearrangement seems to represent an important step in the differentiation of the Hypostomus sp. sex chromosome system. This looks as an uncommon situation among the ZZ/ZW cases so far known in fish, where an initial heterochromatinization seems to have had a more preponderant role.

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References


