Karyotypes of three species of marine catfishes from Brazil

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Abstract: The chromosomes of three species of fishes belonging to the family Ariidae - Netuma barba, Genidens genidens and Arius parkeri - were studied after conventional Giemsa staining. All three species have a diploid chromosome number of $2n = 56$. The karyotype comprises 18 metacentric (m), 18 submetacentric (sm), 18 subtelocentric (st) and 2 telocentric (t) pairs for *N. barba*; 12 m, 20 sm, 20 st and 4 t pairs for *G. genidens*; 16 m, 16 sm, 22 st and 2 t pairs for *A. parkeri*. The results obtained from these three species are compared with each other and with those found in literature.

Resumo: Os cromossomos de 3 espécies de peixes da família Ariidae - *Netuna barba*, *Genidens genidens* e *Arius parkeri* - foram estudados após coloração convencional com Giemsa. As três espécies apresentaram número modal diplóide de $2n = 56$. O cariótipo de *N. barba* compreende 18 pares de cromossomos metacêntricos (m), 18 submetacêntricos (sm), 18 subtelocêntricos (st) e 2 telocêntricos (t); o de *G. genidens* compreende 12 pares m, 20 sm, 20 st e 4 t; o de *A. parkeri* compreende 16 m, 16 sm, 22 st e 2 t. Os resultados obtidos são comparados entre si e com os de outros arídeos encontrados na literatura.

Descriptors: Chromosome, karyotype, Siluriformes, Ariidae.

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Introduction

There are about 2000 species of catfishes belonging to 31 families of which 13 are endemic to South America (Lauder & Liem, 1983). Two of the catfish families became secondarily marine: the Plotosidae (eeltail catfishes) and the Ariidae (marine catfishes) (Kramer, 1990). Members of Ariidae have circumtropical distribution and can be found along all the Brazilian coast. They are of great economical importance in certain regions, mainly for the Brazilian southern sea-side populations (Figueiredo & Menezes, 1978). Numerous studies about this group covering various fields of scientific research have been developed; nevertheless, the systematics and the relationship between its members are still highly controversial (Higuchi et al., 1982; Suzuki & Phan, 1990a).

Cytogenetical investigation has been used as one important tool that can help to elucidate problems related to systematics, evolution (Denton, 1973; Kirpichnikov, 1981) and population structure (e.g. Cestari & Galetti Jr., 1992) of fishes. Karyotypes of only 129 species of freshwater and marine Siluriformes belonging to 13 families are available (Fitzsimons et al., 1988). Just five species of the family Ariidae have been karyotyped until now.

This work belongs to a series of studies that aims to verify genetical aspects of the ariid fishes of the estuarine region of Cananéia, using biochemical (Suzuki & Phan, 1990 a,b), immunological (Gomes et al., in preparation) and cytogenetical (Gomes et al., 1990, 1992) methods. In this paper the karyotypes of *Netuna barba* (Lacépède, 1803), *Genidens genidens* (Valenciennes, 1893) and *Arius*
Materials and methods

Specimens of *N. barba*, *G. genidens* and *A. parkeri* were collected in the estuarine region of Cananéia, São Paulo, Brazil (25°01'S; 47°56'W) from May 1981 to November 1986. Identification and denomination of the species were made after Taylor & Menezes (1977) and Figueiredo & Menezes (1978). The specimens were kept alive in tanks with running sea water and were injected intraperitoneally with 0.2ml/100g body weight of 0.5% colchicine in saline solution. Four to six hours after the injection the animals were sacrificed and the kidney was removed for chromosome preparation following the method described by LeGrande & Fitzsimons (1976) with few modifications (Gomes et al., 1990).

The metaphases were photographed, the chromosomes were counted and the karyograms mounted from photographic prints. The size of the chromosomes in micrometers was estimated by measuring on the microscope one easily identifiable chromosome of a set with a micrometric eyepiece and then correlating these measures with those of all chromosomes of the same set taken from a photographic print (Denton, 1973). The arm ratios (long arm/ short arm) were calculated and terminology for centromeric position followed the criteria of Levan et al. (1964). To facilitate comparisons between species the chromosomes were also grouped in msm and stt types following Uyeno et al. (1983). To calculate arm numbers (AN), m (metacentric) and sm (submetacentric) chromosomes were considered as biarmed and st (subtelocentric) and t (telocentric) as uniarmed.

Results

Metaphase plates were obtained from 20 specimens of *N. barba*, 11 males and 9 females, ranging from 181 mm to 293 mm in total length; 12 specimens of *G. genidens*, 4 males and 8 females, ranging from 145 mm to 240 mm in total length and 13 specimens of *A. parkeri*, 6 males and 7 females, ranging from 206 mm to 356 mm in total length.

In relation to *N. barba*, 203 mitotic spreads were examined, 132 for the males and 71 for the females. The cells with diploid number of 2n = 56 chromosomes account for 77.27% of male and 74.65% of female metaphases. The total length of the chromosomes in micrometers (um) taken as an average of 10 selected spreads ranged from 6.29 µm to 1.81 µm. The karyotype consisted of 9 m pairs, 9 sm pairs, 9 st pairs and 1 t pair (Fig. 1, A and B); AN is, therefore, 92. The 19th st pair is heteromorphic for the males and is constituted by the largest chromosome of the complement, averaging 4.13 µm, and by a smaller one with average total length of 2.49 µm (Fig. 1, A).

Regarding to *G. genidens*, 112 mitotic plates were examined, 69 for the males and 43 for the females. The modal diploid number is 2n = 56, representing 79.07% of the total countings for the males and 79.71% for the females. The total length of the chromosomes in micrometers (µm) taken as an average of 10 selected metaphases ranged from 5.18 µm to 1.77 µm. The karyotype consisted of 6 m pairs, 10 sm pairs, 10 st pairs and 2 t pairs (Fig. 2, A and B). The AN is 88. No heteromorphism was observed between males and females.

From the preparations on *A. parkeri* it was obtained 193 mitotic spreads, 77 for the males and 116 for the females. The modal diploid number is 2n = 56, representing 90.90% of the total countings for the males and 87.07% for the females. The total length of the chromosomes in micrometers (µm) taken as an average of 10 spreads ranged from 4.25 µm to 1.54 µm. The karyotype consisted of 8 m pairs, 8 sm pairs, 11 st pairs and 1 t pair (Fig. 3, A and B). The AN is 88. No heteromorphism between males and females was observed in the chromosome plates examined.

Table 1 summarizes the results obtained in this and in other works ariid fishes.

Discussion

Comparisons of chromosome types between different species or even between the same species studied by different authors may present some problems due to possible mistakes in classifying the elements which are at the boundaries of two distinct morphological categories (LeGrande et al., 1984). For instance, the AN estimated for *Ictalurus catus* by LeGrande et al. (op. cit.) was 84 while by Hudson (1976) it was 64 to 68. Due to this kind of difficulties the differences in AN could be less evident between some species and more obvious between others. This may, in part, be responsible for the great differences in arm number between the karyotypes of *Bagre marinus* and *Bagre bagre* reported by Fitzsimons et al. (1988) and by Gomes et al. (1990), respectively. On the other hand, it's also probable that most of the size variabilities of chromosome arms that alters AN among populations or species is natural and results from mechanisms such as deletions or duplications during karyological evolution. Bearing this in mind, useful comparisons can frequently be made.
Examining the Table 1 it can be seen that all the fishes, including the three species presented here, have $2n = 54$ or 56 chromosomes, and $AN = 80$ or more, except $B.\ marinus$ ($AN = 74$). The number $2n = 56 \pm 2$ with high AN (around 80) has been considered by LeGrande (1981) as characteristics of the ancestral stock from where Siluriformes have evolved.

$N.\ barba$, $G.\ genidens$ and $A.\ parkeri$ have few t chromosomes (one to two pairs) that have not been reported for any other arid fish. In some cases, however, this lack of t chromosomes may be attributed to the fact that in describing karyotypes of fishes some authors prefer to group together the st and t types to avoid mistakes and to facilitate comparisons. For instance, observing the illustration of the karyotype of $B.\ marinus$ (Fitzsimons et al., 1988) one can notice that short arms are not distinguishable in some stt chromosomes that could possibly belong to t types.

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**Fig. 1.** Selected karyotype of a male (A) and a female (B) *Netuma barba.*
Fig. 2. Selected karyotype of a male (A) and a female (B) *Genidens genidens*.

*N. barba*, *G. genidens* and *A. parkeri* have 2n = 56. Of these three species, *N. barba* and *G. genidens* are believed to have similar morphological (Higuchi, 1982), biochemical (Suzuki & Phan, 1990 a, b) and immunological characteristics (Gomes et al., in preparation). From the osteological (Higuchi, 1982) and biochemical point of view (Suzuki & Phan, 1990 a, b) *A. parkeri* is not so similar to *N. barba* and *G. genidens* as are these two later species to each other. Of course, more evidences must be gathered before coming to any definite conclusion but with the development of systematic studies, the karyological characteristics here described could be of some help in the establishment of the relationship between ariid fishes.

Analysing the karyotypes of the three species cited above one could notice some similarities. Besides having the same chromosome number and similar number of m and sst types they have other features in common. For instance, the first m pair of their karyotypes is not very big and not very different in size from the subsequent...
ones. The decreasing in size of the m pairs is slightly more gradual in N. barba and G. genidens than in A. parkeri. The sm chromosomes also show some similarities when comparing the karyotypes of the three species. Their first sm pair is big, the others decrease gradually in size and the last sm one is constituted of small chromosomes. Another feature in common is that their first st pair is the biggest of the complement. Banding techniques would be very useful to verify the possible homologies between chromosome pairs.

In spite of having $2n = 56$, B. bagre has a very distinct karyotype when compared to N. barba, G. genidens and A. parkeri, mainly due to the great differences between their number of msm and stt chromosomes (Table 1).

B. bagre and B. marinus belong to the same genus and are similar in many aspects in spite of having very distinct karyotypes (Gomes et al., 1990). This variability in chromosome morphology could be, for instance, a characteristic of the genus. More data must be obtained before formulating any hypothesis.
Table 1. Karyotypes of some arid catfishes. In parentheses, chromosome types grouped together. AN, arm number; m, metacentric; sm, submetacentric; msm, meta-submetacentric; st, subtelocentric; t, telocentric; stt, subtelo-telocentric

<table>
<thead>
<tr>
<th>Species</th>
<th>2n</th>
<th>AN</th>
<th>m</th>
<th>sm</th>
<th>msm</th>
<th>st</th>
<th>t</th>
<th>stt</th>
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<tr>
<td>Bagre bagre</td>
<td>56</td>
<td>106</td>
<td>24</td>
<td>26</td>
<td>(50)</td>
<td>6</td>
<td>(6)</td>
<td></td>
<td>Gomes et al., 1990</td>
</tr>
<tr>
<td>Bagre marinus</td>
<td>54</td>
<td>74</td>
<td>12</td>
<td>8</td>
<td>(20)</td>
<td></td>
<td>34</td>
<td>Fitzsimons et al., 1988</td>
<td></td>
</tr>
<tr>
<td>Arius dussumieri</td>
<td>54</td>
<td>84</td>
<td>12</td>
<td>8</td>
<td>(30)</td>
<td></td>
<td>24</td>
<td>Rishi et al., 1983</td>
<td></td>
</tr>
<tr>
<td>Ariopsis felis</td>
<td>54</td>
<td>80</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>28</td>
<td>LeGrande, 1980</td>
<td></td>
</tr>
<tr>
<td>Cathorops sp</td>
<td>54</td>
<td>80</td>
<td>13</td>
<td>13</td>
<td>(26)</td>
<td>28</td>
<td>(28)</td>
<td></td>
<td>Gomes et al., 1992</td>
</tr>
<tr>
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<td>56</td>
<td>92</td>
<td>18</td>
<td>18</td>
<td>(36)</td>
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<td>2</td>
<td>(20)</td>
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</tr>
<tr>
<td>G. genidens</td>
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<td>88</td>
<td>12</td>
<td>20</td>
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<tr>
<td>A. parkeri</td>
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<td>88</td>
<td>16</td>
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<td>22</td>
<td>2</td>
<td>(24)</td>
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Examining all the arid fishes karyotyped until now, one can notice that Arius dussumieri (Rishi et al., 1983), Ariopsis felis (LeGrande, 1980) and Cathorops sp (Gomes et al., 1992) have karyotypes more similar to one another than to the other species (Table 1). They all have 2n = 54 and similar numbers of msm and stt chromosomes (see Gomes et al., 1990, 1992), and consequently similar AN (84, 80, 80, respectively). In comparison with these species, N. barba, G. genidens and A. parkeri have higher AN due to the greater number of chromosomes and a slightly higher number of msm elements.

Among the arid fishes studied to the moment N. barba was the unique to present chromosome heteromorphism in the male. The configuration of the 19th pair in the male suggests a XX/XY sex chromosome system. Banding techniques would be valuable to confirm this configuration. However, to assess the role of these chromosomes in sex determination, other kind of methods such as crossing and hybridization, must be used. Similar configuration was also found in Siluriformes for Notunus (Rabda) taylori (LeGrande, 1981).

Conclusion

It's impossible to come to definitive conclusions about the karyological evolution of arid catfishes with the few data available until now, but it seems that the continuation of this kind of studies, in addition to morphological, osteological and biochemical ones, can contribute to the understanding of the evolution of the family and the relationship between its members.

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