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SHORT COMMUNICATION

Conservative distribution of 5S rDNA loci in *Schizodon* (Pisces, Anostomidae) chromosomes

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Little is known on 5S rDNA chromosomal localization in the rich neotropical fish fauna (Martins & Galetti 1999). In order to investigate the variability of chromosomal sites of 5S RNA genes in neotropical fishes, chromosomal localization of these genes was performed in six *Schizodon* species collected in different South America hydrographic systems. *Schizodon altoparanae* was collected near to the township of Salto Grande, SP, Brazil (Paranapanema River), *S. borelli* was caught at four collection sites along the Paraguay River Basin (Cuiabá River, Santo Antônio do Laverger, MT; Bento Gomes River, Poconé, MT and Miranda and Vermelho Rivers, Passo do Lontra, Corumbá, MS, Brazil), *S. isognathum* was collected in the Vermelho River and *S. nasutus* came from two localities: the Mogi-Guaçu River (township of Pirassununga, SP, Brazil) and the Paraná River (Missiones, Argentina). *S. knerii* was collected in the municipality of Três Marias, MG, Brazil (São Francisco River) and finally *S. vittatus* was caught in the Araguaia River near the township of Barra do Garças, MT (Amazon Basin).

Mitotic chromosomes were obtained from anterior kidney cells following a conventional air drying technique described elsewhere. The recombinant plasmid pBSIIKS containing the 5S rRNA gene obtained from *Leporinus elongatus* (Martins & Galetti 1999, Martins & Galetti in preparation) was used as a probe in fluorescence *in-situ* hybridization (FISH). A basic FISH protocol and the sequential identification of NORs (nucleolus organizer regions) were carried out as described in Martins & Galetti (1998, 1999).

In agreement with previous studies (Martins & Galetti 1997, 1998), the karyotype of all *Schizodon* showed $2n = 54$ biarmed chromosomes. FISH detected a major 5S cluster located subterminally in a medium-sized metacentric chromosomal pair (14th pair) of all studied species (Figure 1). Additionally, a minor 5S cluster was detected near to the centromere area on the long arm of a medium-sized submetacentric pair (12th pair). NOR sites (45S rRNA gene sites), confirmed by sequential Ag-staining of 5S FISH slides, were identified in a small metacentric pair (20th pair) for all *Schizodon* investigated (Figure 1a), as previously reported (Martins & Galetti 1997, 1998). Such divergent locations of NORs and 5S rDNA loci seem to be the most common situation observed in fish (Martínez *et al.* 1996) and by far the most frequent distribution pattern observed in vertebrates (Suzuki *et al.* 1996), although the 5S

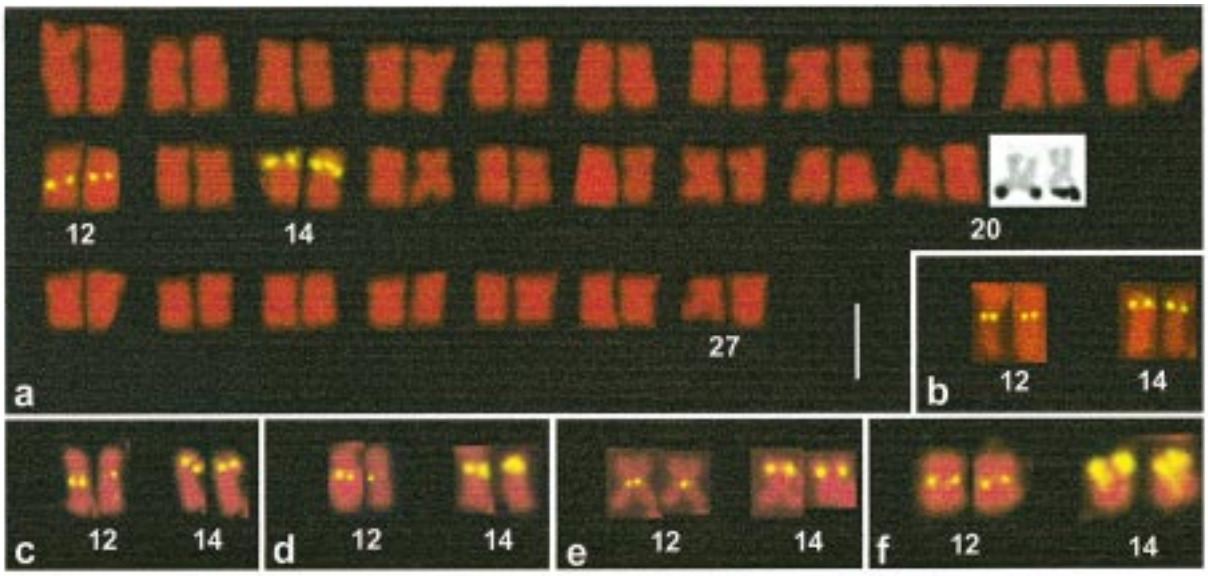

Figure 1. 5S-FISH karyotype of *Schizodon vittatus* (**a**) and chromosome pairs bearing 5S rDNA sites in *S. altoparanae* (**b**), *S. knerii* (**c**), *S. borelli* (**d**), *S. isognathum* (**e**) and *S. nasutus* (**f**). The 5S rDNA is located (yellow signals) in the chromosomal pairs 12 and 14 in all species. The white box (pair 20) in (**a**) shows the Ag-NOR bearing pair. Scale bar = 4 μm.

and 45S rDNA loci were linked in the same chromosomal pair in *Salmo salar* (Pendás *et al.* 1994). In eukaryotes the large 45S rRNA genes are transcribed by the nucleolar enzyme RNA polymerase I, while the 5S genes are transcribed far from the nucleolus by the non-nucleolar RNA polymerase III. It is suggested that such functional divergences would require different physical location between the large rDNA and the 5S arrays. In addition, gene conversion and unequal crossing-over could be important processes in the maintenance of a conserved sequence in multiple tandem arrays (Dover 1986). These mechanisms might be more efficient regardless of whether the 5S and 45S clusters remain separated from each other instead of in a linked configuration, avoiding disruptive interferences, such as undesired translocation of 5S sequences inside the 45S arrays (Martins & Galetti 1999), and explaining why most vertebrates have these clusters on distinct chromosomes.

Multiple loci for 5S rDNA genes have been detected among several fish (Fujiwara *et al.* 1998) and other vertebrates (Lucchini *et al.* 1993). A dual system represented by oocyte and somatic types of 5S genes, which are regulated differently in oocytes and somatic cells, has been detected in several vertebrates, including fish and amphibian (Komiya *et al.* 1986). Major and minor 5S rDNA sites have also been described in humans, monkeys and other mammals (Suzuki *et al.* 1996). Moreover, the occurrence of 5S gene arrays located in more than one chromosomal pair, such as in *Schizodon*, suggests that these rDNA clusters can be evolving separately and could represent distinct 5S rDNA units. Similar 5S chromosome sites were previously reported in another Anostomidae genus *Leporinus* (Martins & Galetti 1999). It is probable that these major and minor 5S clusters occupy homeologous chromosomes in both genera, and that the multiplicity of 5S loci is ancient among Anostomidae.

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