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Occurrence of *Akodon cursor* (Rodentia, Cricetidae) with 14, 15 and 16 chromosome cytotypes in the same geographic area in Southern Brazil

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ABSTRACT

The karyotype of *Akodon cursor* (initially identified as *A. arviculoides*) had been reported with chromosomal numbers 14 and 15 in the South and Southeast and 16 in Northeastern Brazil. We found the three cytotypes in a region of Southern Brazil. The G-band patterns of these specimens were the same as those from southeastern and northeastern regions. Seventeen different combinations of chromosomes due to a complex rearrangement in pair 1 and pericentric inversions in pairs 2 and 3 were identified. Seven of these combinations are new to in the literature.

INTRODUCTION

Akodon is widely distributed throughout South America, except for the lowlands of the Amazon basin (Myers and Patton, 1989). This genus includes species very similar in morphology, some of them difficult to separate taxonomically, although there is considerable karyotypic diversity (2n = 14 to 52). The intraspecific and intrapopulational chromosomal variations are due to structural and numerical rearrangements. Centric fusion in *Akodon* has been found by Bianchi *et al.* (1969, 1971, 1973) in *A. molinae* (2n = 42 to 44); Bianchi *et al.* (1971, 1979) and Kiblisky *et al.* (1976) in *A. dolores* (2n = 34 to 40); and Yonenaga-Yassuda *et al.* (1987) in *A. reinhardti* (2n = 37, 38).

In *A. cursor* the diploid numbers 14 and 15 have been found in Paraná (Bossle *et al.*, 1988), São Paulo (Yonenaga, 1972; Yonenaga *et al.*, 1975 and Yonenaga-Yassuda *et al.*, 1983) and Rio de Janeiro (Yonenaga-Yassuda, 1979) and 16 in Pernambuco (Maia and Langguth, 1981). These differences are due to complex rearrangements comprising centric fission-fusion and pericentic and paracentric inversions. Changes in the number of autosomal arms produced by pericentric inversions occur in three autosomal pairs (Yonenaga, 1972; Yonenaga *et al.*, 1975; Yonenaga-Yassuda, 1979; Maia and Langguth, 1981; Yonenaga-Yassuda *et al.*, 1983; Bossle *et al.*, 1988).

MATERIAL AND METHODS

A total of 97 specimens of *A. cursor* (55 males and 42 females) were collected from different localities of Guaraqueçaba Bay in Paraná State, Southern Brazil (25°15'S and 48°30'W). They were taken from four islands (Rabelo, Rasa, Laranjeiras and Gamelas) and three localities on the mainland (Tromomô, Laranjeiras and Massarapuã) (Figure 1a). The specimens were

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prepared as standard museum skins and skulls and the voucher specimens are in the collection of the Departamento de Genética, Universidade Federal do Paraná, Curitiba, Brazil. Chromosomes were obtained from bone marrow by using Ford and Hamerton's (1956) technique. G-banding was obtained by trypsin treatment (Seabright, 1971, with modifications). Chromosome nomenclature used in this paper is that used by Yonenaga-Yassuda (1979).

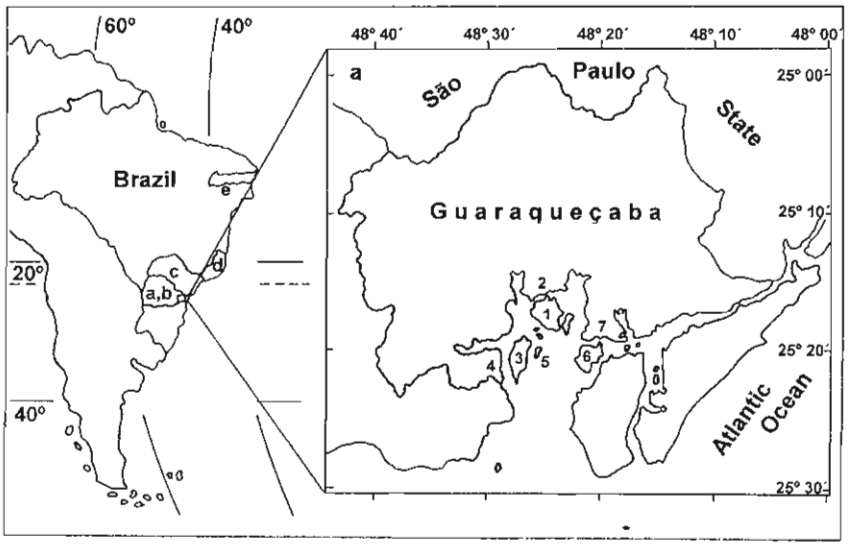

Figure 1 - Map of South America showing Brazilian distribution of karyotypic forms of *Akodon cursor*: a) Paraná - Guaraqueçaba with capture points studied in this investigation: 1, Rabelo island (2n = 14, 15); 2, Tromomô (2n = 14, 15, 16); 3, Rasa island (2n = 14, 15, 16); 4, Massarapuã (2n = 14, 15, 16); 5, Gamelas island (2n = 14, 15); 6, Laranjeiras island (2n = 14, 15); 7, Laranjeiras mainland (2n = 14, 15); b) Paraná - Serra do Mar (2n = 14, 15); c) São Paulo (2n = 14, 15); d) Rio de Janeiro (2n = 14, 15); e) Pernambuco (2n = 16).

RESULTS

Forty-nine animals (29 males and 20 females) had 2n = 14 and NA (number of autosomal arms) ranging from 18 to 21; 22 males and 22 females had 2n = 15 and NA = 20 to 24, and four males had 2n = 16 and NA = 23, 24. The 2n = 14 forms (Figure 2A) had a pair 1 with two large metacentric chromosomes; pair 2 comprised two medium-sized submetacentrics and/or acrocentrics; pair 3 had medium-sized acrocentrics and/or metacentrics; pair 4 were medium-sized metacentrics; pair 5, medium-sized acrocentrics, and pair 6, minute metacentrics. The sex-chromosome pair had two acrocentric homomorphic Xs in females, and two heteromorphic acrocentrics in XY males; the Y chromosome being similar in size to pair 6.

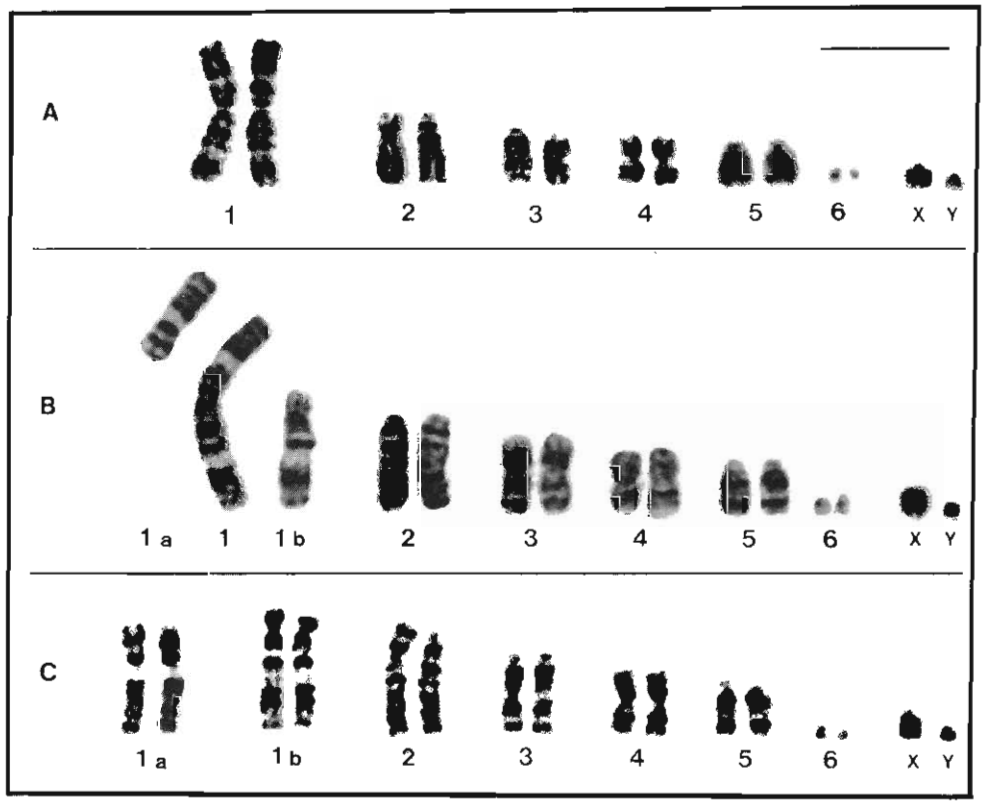

Figure 2 - G-band patterns of cytotypes of *Akodon cursor*. A) 2n = 14, NA = 19, chromosome 1 in homozygosis, pair 2 heterozygous, pair 3 homozygous for acrocentric form; B) 2n = 15, NA = 22, the unique chromosomes 1, 1a and 1b, pair 2 homozygous for acrocentric form, pair 3 homozygous for metacentric form; C) 2n = 16, NA = 23, chromosomes 1a and 1b in homozygosis, pair 2 heterozygous, pair 3 homozygous for acrocentric form. Bar = 10 µm.

Chromosomal polymorphisms were detected in pairs 2 and 3 and were due to pericentric inversions. The 21 different karyotypes so far reported in *A. cursor* are shown in Table I. Seventeen were found in Guaraqueçaba, seven of which had not been previously reported by other authors. The ten karyotypes found in Guaraqueçaba and in other regions were: A, B, C, D, E, G, I, J, L, and M. The seven karyotypes found only in Guaraqueçaba were: F, N, O, P, Q, R, and S. In pair 2, we detected 53% of specimens homozygous for the acrocentric form, 43%

Table I - Karyotypes described for *Akodon cursor*.

Karyotype	2n	NA	Pair[1] 2	Pair[1] 3	Specimens studied[2]	Locality[3] (States)	References
A	14	18	A	A	16 (2; 3)	SP, RJ, PR	Yonenaga, 1972; Yonenaga *et al.*, 1975; Yonenaga-Yassuda, 1979; Yonenaga-Yassuda *et al.*, 1983; Fagundes, 1993; this study
B	14	19	A	H	45 (9; 8)	SP, RJ, PR	Yonenaga, 1972; Yonenaga *et al.*, 1975; Yonenaga-Yassuda, 1979; Yonenaga-Yassuda *et al.*, 1983; Fagundes, 1993; this study
C	14	19	H	A	12 (2; 1)	SP, RJ, PR	Yonenaga, 1972; Yonenaga *et al.*, 1975; Yonenaga-Yassuda, 1979; Yonenaga-Yassuda *et al.*, 1983; Fagundes, 1993; this study
D	14	20	A	M	13 (2; 3)	SP, PR	Yonenaga, 1972; Yonenaga *et al.*, 1975; Fagundes, 1993; this study
E	14	20	H	H	18 (4; 4)	SP, RJ, PR	Yonenaga *et al.*, 1975; Yonenaga-Yassuda, 1979; Yonenaga-Yassuda *et al.*, 1983; Fagundes, 1993; this study
F	14	20	SM	A	2 (1; 1)	PR	This study
G	14	21	H	M	10 (9 males)	SP, PR	Yonenaga-Yassuda *et al.*, 1983; this study
H	14	21	SM	H	1	SP	Yonenaga *et al.*, 1975
I	15	20	A	A	6 (2; 3)	SP, PR	Fagundes, 1993; this study
J	15	21	A	H	12 (4; 6)	SP, PR	Yonenaga-Yassuda, 1979; Fagundes, 1993; this study
K	15	21	H	A	1	SP	Fagundes, 1993
L	15	22	A	M	9 (5; 3)	SP, PR	Yonenaga-Yassuda, 1979; this study
M	15	22	H	H	15 (7; 7)	SP, PR	Yonenaga-Yassuda, 1979; this study
N	15	23	H	M	5 (3; 2)	PR	This study
O	15	23	SM	H	1 (male)	PR	This study
P	15	24	SM	M	1 (female)	PR	This study
Q	16	23	A	H	1 (male)	PR	This study
R	16	23	H	A	2 (males)	PR	This study
S	16	24	H	H	1 (male)	PR	This study
T	16	25	SM	H	2	PE	Maia and Langguth, 1981
U	16	26	SM	M	23	PE	Maia and Langguth, 1981

NA = Number of autosomal arms.
[1]A = Acrocentric; H = heterozygous; M = metacentric; SM = submetacentric.
[2]The number of males and females, respectively, from Guaraqueçaba (PR) are shown within parentheses.
[3]SP = São Paulo; RJ = Rio de Janeiro; PR = Paraná; PE = Pernambuco.

heterozygous, and 4% homozygous for the submetacentric form. Pair 3 showed 17% homozygous for acrocentrics, 54% heterozygous, and 29% metacentrics in homozygosis. Both pairs were in Hardy-Weinberg equilibrium (pair 2: $\chi^2_{(2)} = 0.0174$; pair 3: $\chi^2_{(2)} = 0.0075$; $P > 0.99$).

All kinds of possible combinations considering pairs 2 and 3 were found in Guaraqueçaba (PR). The homozygous acrocentric pair 2 associated with heterozygous pair 3 were the most frequent combination. Homozygous for submetacentric in pair 2 with homozygous for metacentric or heterozygous in pair 3 were the two rarest combinations.

The numerical differences among the three cytotypes are due to a complex rearrangement involving centric fusion-fission in the chromosomes of pair 1. Specimens with 2n = 14 (Figure 2A) showed two metacentric chromosomes 1. In 2n = 15 (Figure 2B) there are three elements: a large metacentric (1) and two different submetacentrics (1a and 1b), while in 2n = 16 (Figure 2C) pair 1 is substituted by pairs 1a and 1b in the homozygous state. G-banding shows that 1a and 1b

have homology with the short and long arms of pair 1, and G-band patterns also show an inversion when 1a is compared to the short arm of chromosome 1 (Figure 2B). In three of the seven localities studied (Figure 1a) (Rasa island, Tromomô and Massarapuã) we observed the three cytotypes.

DISCUSSION

Comparative analysis of G-band patterns among the three cytotypes found in this study with those described for southern (Figure 1b; Bossle *et al.*, 1988), southeastern (Figure 1c; Yonenaga, 1972 and Figure 1d; Yonenaga-Yassuda, 1979) and northeastern regions (Figure 1e; Maia and Langguth, 1981) shows homologies among all chromosomal pairs.

Cytogenetic studies of *A. cursor* from different regions of Brazil (Table I) demonstrate geographic variation in diploid number and a remarkable chromosomal polymorphism, due to a high frequency of pericentric inversions involving pairs 2 and 3. Chromosomal variation in pair 5 was observed only in a female from Rio de Janeiro State studied by Yonenaga-Yassuda (1979). The high frequency of heterozygosity for pairs 2 and 3 was also reported by Yonenaga (1972), Yonenaga *et al.* (1975), Yonenaga-Yassuda (1979), Yonenaga-Yassuda *et al.* (1983), Bossle *et al.* (1988), and Fagundes (1993). Maia and Langguth (1981) studied specimens with 2n = 16 from Pernambuco State. They did not find the acrocentric chromosome form in pair 2. Twenty-seven karyotypes are expected, considering the possible combinations among all chromosome forms of pairs 1, 2, and 3. As the pericentric inversion in pair 5 is unusual it was disregarded as a possible source of karyotype variation. Twenty-one karyotypes for *A. cursor* have been described so far, and six karyotypes remain to be found. The polymorphisms in pairs 2 and 3 of our sample were in Hardy-Weinberg equilibrium. This high frequency of heterozygotes does not seem to reduce fertility in this species.

White (1978) postulated that chromosomal rearrangements play an important role in initiating divergence during speciation. They may reduce the viability of embryos or, if they do not depress viability, they may produce meiotic arrest. King (1987, 1993) considers chromosomal rearrangements as post-mating isolating mechanisms; yet this author proposes that different kinds of events may occur in the meiosis of some Rodentia species, nullifying the deletereous effect of heterozygosis for pericentric inversions. Centric fusion and pericentric inversion are potentially negative in heterozygous specimens. Once the aneuploid gametes are produced, rearrangements could even decrease the viability of the individuals. However, this seems not to occur because many heterozygotes can be observed in *A. cursor* from Guaraqueçaba and other localities. Fagundes (1993) found *A. cursor* males of São Paulo State with normal meiosis, probably due to the existence of heterosynapsis in regions with pericentric inversion. This can represent a heterotic condition, with increased fitness of the heterozygotes compared to homozygotes. For White (1978), both heterosis and frequency-dependent selection could lead to the polymorphic state of a rearrangement. White (1978) also suggested that chromosomal rearrangements existing in a balanced polymorphic state are more likely to be cohesive than divisive agents in natural populations. King (1993) argue that chromosomal polymorphisms with a high frequency of heterozygosis - as we observe in *A. cursor* - could not be associated with speciation.

In the south (Bossle *et al.*, 1988) and southeast (Yonenaga, 1972 and Yonenaga-Yassuda, 1979) specimens with 14 and 15 chromosomes are frequent while 2n = 16 specimens are found in the northeast (Maia and Langguth, 1981). We found specimens with 14, 15 and 16 chromosomes in the same geographic area in three out of seven localities of Guaraqueçaba Bay. Moreover, in our series pericentric inversions were only found in pairs 2 and 3. Bossle *et al.* (1988) in a study of seven specimens (2n = 14, 15) collected from Serra do Mar (Antonina City, Paraná State) also observed pericentric inversions only in chromosomes 2 and 3.

Sbalqueiro, I.J. and Nascimento, A.P., unpublished results, were able to obtain specimens under laboratory conditions with 14, 15 and 16 chromosomes from crosses of specimens with 2n = 15. This supports the assumption that a similar phenomenon may occur in wild populations of *A. cursor*.

The submetacentric form in pair 2 was found in all places where the species was studied, but the acrocentric form was not observed in Pernambuco. The heterozygosity of pair 3 was detected in specimens from all the localities so far assessed; however, acrocentric and metacentric homozygous forms have not yet been found in Pernambuco and Rio de Janeiro, respectively.

According to Yonenaga-Yassuda (1979) the polymorphism detected in pair 1 of this species can be explained by two alternative mechanisms: a) pericentric inversions in submetacentrics 1a and 1b followed by centric fusion or b) chromosomal dissociation of the largest metacentric followed by pericentric inversion in the segment corresponding to the long arm, originating 1b chromosome, and centromeric activation followed by paracentric inversion in the other segment, originating 1a.

Alfredo Langguth (personal communication in Fagundes, 1993) considers the northeastern specimens as a different species of *A. cursor*, suggesting the temporary denomination of *A.* aff. *cursor*. The occurrence of 2n = 16 in Guaraqueçaba does not support this suggestion, although it is not sufficient to discard it.

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RESUMO

O cariótipo de *Akodon cursor* (inicialmente identificado como *A. arviculoides*) tinha sido relatado com números cromossômicos 14 e 15 no Sul e Sudeste do Brasil e 16 no Nordeste brasileiro. Neste trabalho é mostrado, pela primeira vez, a ocorrência conjunta das três formas cariotípicas, em três locais de coleta, na baía de Guaraqueçaba, região Sul do Brasil. Os padrões de bandamento G do complemento cromossômico dos espécimens do Sul não mostram diferenças com relação aos padrões das regiões Sudeste e Nordeste. Foram identificadas 17 combinações cariotípicas diferentes devido a um rearranjo complexo no par 1 e a inversões pericêntricas nos pares 2 e 3. Sete destas combinações ainda não tinham sido relatadas na literatura.

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