

A new karyotype of *Oligoryzomys* (Sigmodontinae, Rodentia) from central Brazil

J. FERNANDO DE S. LIMA¹, CIBELE R. BONVICINO² and SANAE KASAHARA³

¹ Departamento de Genética, Bolsista do CNPq, Universidade Federal do Paraná, Curitiba, PR, Brazil

² Divisão de Genética, Instituto Nacional de Câncer, Praça Cruz Vermelha, and Departamento de Medicina Tropical, FIOCRUZ, Brazil

³ Departamento de Biologia, IB, Universidade Estadual Paulista, Rio Claro, SP, Brazil

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A new karyotype of the genus *Oligoryzomys* was described for specimens collected in Brazilian Cerrado in Tocantins and Goiás States. Conventional staining, G-, C-banding, and Ag-NOR staining techniques were used for describe this karyotype with $2n = 70$, $AN = 74$ or 76 . The chromosome complement, with the highest diploid number known among *Oligoryzomys* species, differs from all others previously reported. This small sized *Oligoryzomys* species can be differentiated from other Brazilian *Oligoryzomys* not only by chromosomal complement, but also in some morphological attributes. The new species is apparently restricted in distribution and endemic of Brazilian Cerrado, occurring only in Rio Tocantins basin.

J. Fernando De S. Lima, UNESP/FMVZ, Depto de Produção e Exploração Animal, Sector de Animais Silvestres, C. P. 560, C. E. P. 18.618-000, Botucatu – SP, Brazil. E-mail: jfslim@hotmail.com

The genus *Oligoryzomys* Bangs, 1900 has a wide-spread distribution, from northern Central America to the southern part of South America. Fifteen species were recognized in the latest checklist of the genus, seven of which occurring in Brazil: *O. chacoensis*, *O. delticola*, *O. eliurus*, *O. flavescens*, *O. fulvescens*, *O. microtis*, and *O. nigripes* (MUSSEY and CARLETON 1993). These *Oligoryzomys* species occur in four Brazilian biomes: Cerrado, Caatinga, Atlantic Forest and Amazon. Recently, *O. stramineus*, an endemic species of the Brazilian Cerrado and Caatinga was described (BONVICINO and WEKSLER 1998) while *O. fornesi* Massoia, 1973 was recognized as a valid species (MYERS et al. 1995; BONVICINO and WEKSLER 1998). These findings extended the number of Brazilian *Oligoryzomys* species to nine. However, previous studies based on morphologic similarities and G-band pattern suggested that *O. nigripes* was as senior synonymous of *O. delticola* and *O. eliurus* (BONVICINO and WEKSLER 1998).

Oligoryzomys karyology, morphology, and geographic distribution have been reported in the last 25 years by many authors (GARDNER and PATTON 1976; ALMEIDA and YONENAGA-YASSUDA 1991; ESPINOSA and REIG 1991; SBALQUEIRO et al. 1991; BONVICINO et al. 2001). Other inventories allow the identification of new species and karyotypes of *Oligoryzomys* in Brazil (SILVA and YONENAGA-YASSUDA 1997; BONVICINO and WEKSLER 1998). Due to the extreme similarity in morphological attributes among

Oligoryzomys species, karyologic studies have been used to clarify the taxonomy of some species and understanding the diversification of the genus. The highest diploid number in the genus $2n = 68$, $AN = 74$ or 76 was found in Peru (GARDNER and PATTON 1976) and the lowest diploid numbers, $2n = 44$, $44/45$, $AN = 52$, $52/53$ in Brazil (SILVA and YONENAGA-YASSUDA 1997).

In this paper, we describe a new *Oligoryzomys* karyotype, with the highest diploid number in the genus, comment on the geographic distribution and morphological characteristics of this karyomorph, herein referred to as *Oligoryzomys* sp., and compare our findings with karyologic data on related species.

MATERIAL AND METHODS

We karyotyped 6 specimens of *Oligoryzomys* collected in the following Brazilian localities (Fig. 1: 1, Fazenda Elizeu ($9^{\circ}55'S$, $48^{\circ}17'W$), Lajeado, Tocantins State: female MN LJ35; 2, Chácara União (around $10^{\circ}44'S$, $48^{\circ}23'W$), Porto Nacional, Tocantins State: female ZUT 30; 3, Fazenda Fiandeiras, Cavalcante ($13^{\circ}47'51''S$, $47^{\circ}27'30''W$), Chapada dos Veadeiros National Park, Goiás State: females MN 50320, 50321, male MN 50318; 4, Fazenda Cadoz, Mimoso de Goiás ($15^{\circ}03'22''S$, $48^{\circ}09'41''W$), Goiás State: female MN 67087.

Chromosome preparations were obtained in the field from bone marrow cells, after in vivo colchicine

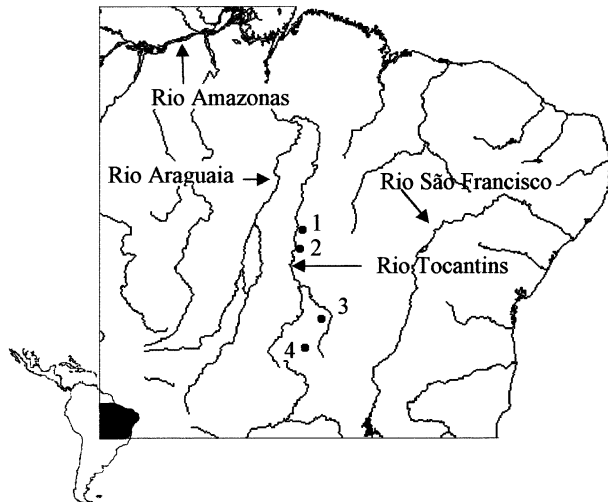


Fig. 1. Brazilian localities of occurrence of *Oligoryzomys* sp. with $2n = 70$. Tocantins State: (1) Lajeado and (2) Porto Nacional; Goiás State: (3) Cavalcante and (4) Mimoso de Goiás.

treatment, or from short-term bone marrow cultures in RPMI 1640 (80 %), foetal bovine serum (20 %), colchicine (10^{-6} M) and ethidium bromide ($5\mu\text{g ml}^{-1}$) for 2 hours (BONVICINO et al. 2001). G- and C- banding were obtained in specimens from localities 1 and 2 following SEABRIGHT (1971) and SUMNER (1972), respectively. Ag-NOR staining was

carried out following HOWELL and BLACK (1980). Skins and skull of specimens were deposited in the mammal collection of Museu Nacional (MN, UFRJ), Rio de Janeiro, Brazil, and in the mammal collection of Fundação Universidade do Tocantins (ZUT, UNITINS), Porto Nacional, Tocantins State, Brazil.

RESULTS

Karyological analyses of the two specimens from localities 1 and 2 showed $2n = 70$, $AN = 76$ (Fig. 2). The autosome complement is composed by 30 acrocentric pairs varying in size from large to small and 4 biarmed pairs varying in size from medium to small, the X chromosome is a large submetacentric, the greatest of the chromosome complement. The G banding allowed the identification of the chromosome pair (Fig. 3). Karyological analyses of four specimens from localities 3 and 4 showed $2n = 70$, $AN = 74$. The autosome complement comprises 31 acrocentric pairs (one large pair and 30 pairs varying in size from medium to small) and 3 pairs of small sized biarmed chromosomes. The X chromosome is a large submetacentric, and the Y chromosome a small acrocentric.

The C- banding showed pericentromeric heterochromatin in almost all autosome pairs, except the

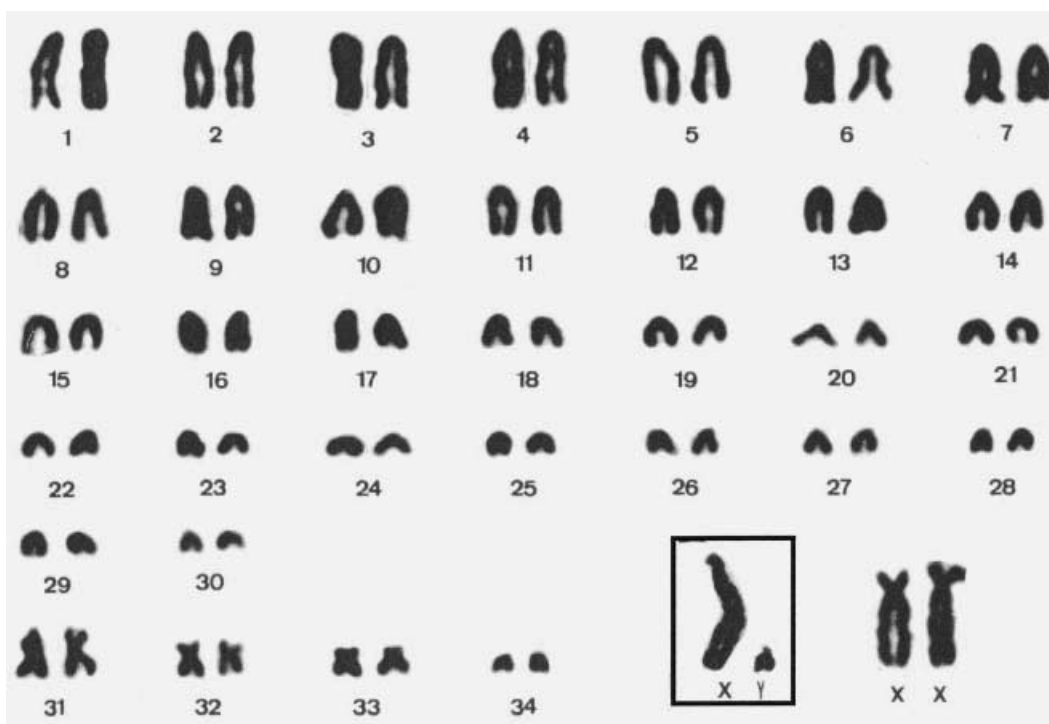


Fig. 2. Conventional Giemsa staining karyotype of *Oligoryzomys* sp., female ZUT 30 ($2n = 70$, $AN = 76$). Inset: the sexual pair of the male.

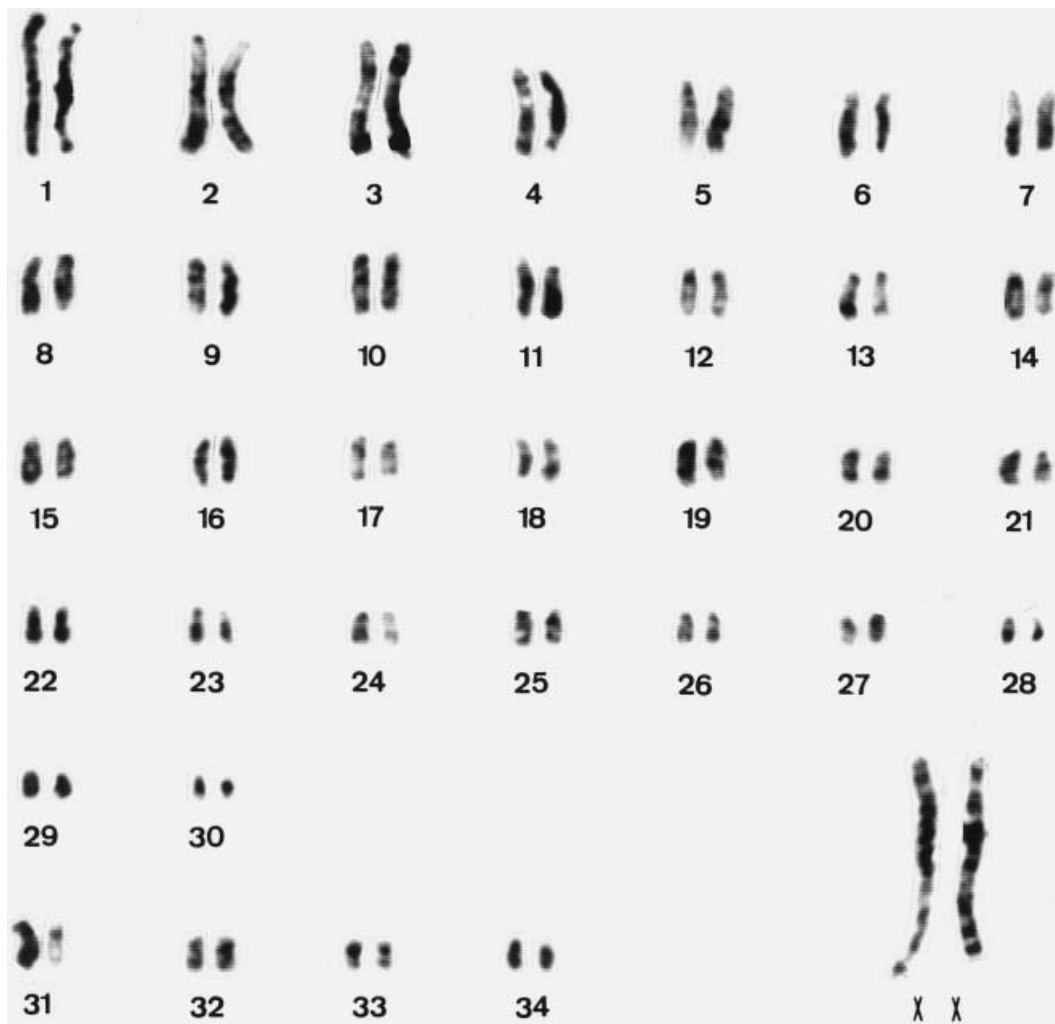


Fig. 3. G- banded karyotype of *Oligoryzomys* sp., female MN LJ35 ($2n = 70$, $AN = 76$)

pair 34; the short arm of X chromosome is almost completely heterochromatic (Fig. 4). Ag-NORs were located in the telomeric region of the short arms of acrocentric chromosomes of large and medium size, and their number in each metaphase varies from 3 to 7, being 4 the modal number (Fig. 5).

Morphological comparisons allow the differentiation of the specimens of *Oligoryzomys* of the present sample in the following external characters: (1) a creamy ventral coloration and an undefined limit between dorsal and ventral colour against a whitish venter and a definite limit between dorsal and ventral coloration in adult specimens of *O. nigripes* and *O. stramineus*; (2) total absence of yellow hair at ventral region between forelimbs against presence of a yellow patch in *O. nigripes* and *O. stramineus*; (3) bicolor tail against tail with only one predominant color in *O. nigripes*; (4) completely creamy ventral side of limbs against dark coloration in *O. fornesi* and *O. flavescens*; (5) lighter dorsal and heterogeneous col-

oration (lined with dark and light hairs) against darker and homogeneous coloration in *O. flavescens*.

DISCUSSION

The specimens of *Oligoryzomys* sp. differed from all other *Oligoryzomys* species in the chromosome complement and morphology. As previously defined, the Brazilian *Oligoryzomys* species can be divided into two groups, the small body-size and the large body-size (BONVICINO and WEKSLER 1998). All small body-size Brazilian *Oligoryzomys* species have allopatric distributions whereas, among large body-size species, the distributions might be sympatric. The karyotypes of species belonging to the small body sized group (*O. fornesi*, *O. microtis*, *O. flavescens*, and *Oligoryzomys* sp.) shared some similarities. All of them have a predominance of acrocentric chromosomes and a variable number of few small biarmed elements, two pairs in *O. microtis*, *O. flavescens* (with

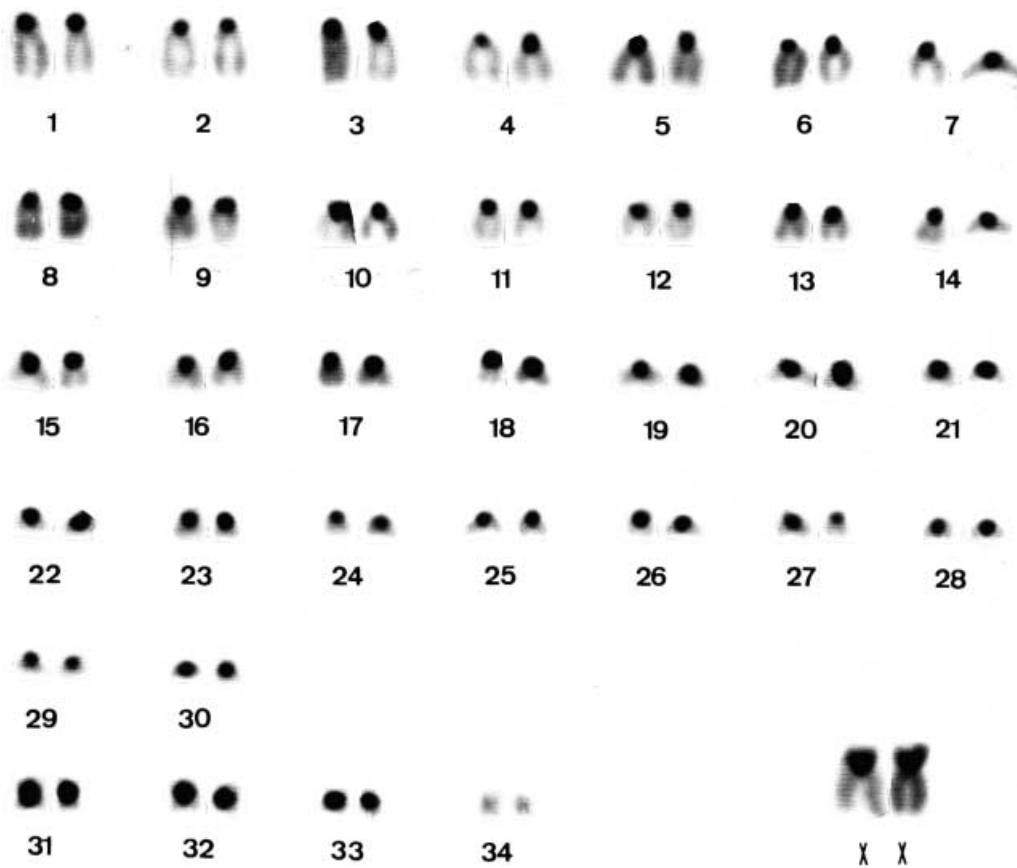


Fig. 4. C banded karyotype of *Oligoryzomys* sp., female ZUT 30 ($2n = 70$, $AN = 76$).

biarmed B chromosomes) and *O. fornesi*, and three or four pairs in *Oligoryzomys* sp. On the other hand, the species belonging to the large body sized group (*O. nigripes*, *O. chacoensis* and *O. stramineus*) shared a high fundamental number in respect to diploid number, due to more numerous biarmed chromosomes in the karyotype.

The two karyotypes here described shared the same diploid number ($2n = 70$) but differ in fundamental number ($AN = 74$ and 76). This difference is probably due to a pericentric inversion affecting a medium sized pair, biarmed in localities 1 and 2 acrocentric in localities 3 and 4. This chromosome rearrangement could not be confirmed because G-banding pattern was not available for specimens from localities 3 and 4. The karyotype of *Oligoryzomys* sp. showed the highest diploid number in the genus and differ from all other previously reported (Table 1). The $2n = 70$, $AN = 74$ or 76 karyotypes of *Oligoryzomys* sp. are more similar to that of *O. longicaudatus* variant 1 with $2n = 68$, $AN = 74$ or 76 (GARDNER and PATTON 1976) than those found in any other *Oligoryzomys* species. They differed only in the number of acrocentrics, which is 30 or 31 in the $2n = 70$ karyotypes and 28 or 29 in the $2n = 68$ karyotypes. The

karyotype of *Oligoryzomys* sp. showed strongly stained pericentromeric heterochromatin in the majority of autosomes pairs, similar to the pattern found in some other congeneric species (SBALQUEIRO et al. 1991; ESPINOSA and REIG 1991; ANISKIN and VOLOBOUV 1999). *O. nigripes* (ALMEIDA and YONE-NAGA-YASSUDA 1991) is one exception, presenting less stained small blocks of heterochromatin in the autosomes. In the karyotype here analysed, the Ag-NORs are restricted to the telomeric region of the short arms of medium and large sized acrocentric chromosomes, similar to the pattern found in other *Oligoryzomys* species (FURTADO 1981, SVARTMAN 1989; SBALQUEIRO et al. 1991; SILVA and YONE-NAGA-YASSUDA 1997).

Karyologic and morphologic data strongly suggest that the karyomorphotype here analysed is a new undescribed species, apparently endemic in Brazilian



Fig. 5. Ag-NOR bearing chromosomes of *Oligoryzomys* sp., female ZUT 30 ($2n = 70$, $AN = 76$).

Table 1. Data on *Oligoryzomys* karyotypes. Taxa designated the valid species name, sometimes different from the names cited in original references. References: 1. Brum-Zorrilla et al. (1988), 2. Sbalqueiro et al. (1991), 3. Furtado (1981), 4. Yonenaga et al. (1976), 5. Bonvicino et al. (2001), 6. Gardner and Patton (1976), 7. Haiduk et al. (1979), 8. Espinosa and Reig (1991), 9. Bonvicino and Weksler (1998), 10. Zanchin (1988), 11. Silva and Yonenaga-Yassuda (1997), 12. Svartman (1989), 13. Almeida and Yonenaga-Yassuda (1991), 14. Myers and Carleton (1981).

Taxa	2n	NA	Locality	Reference
<i>O. andinus</i>	60	70	Peru	6
<i>O. chacoensis</i>	58	74	Argentina; Paraguay	8, 14
<i>O. delticola</i>	62	80–82	Brazil: RS, PR; Argentina; Uruguay	1, 2, 8
<i>O. destructor</i>	60	76	Peru; Uruguay	1
<i>O. flavescens</i>	64	68	Brazil: MG,	9
<i>O. flavescens</i>	66	70	Argentina; Uruguay	1
<i>O. flavescens</i>	64–66	66–68		1, 2, 8
<i>O. flavescens</i>	66–68	68–70	Argentina	8
<i>O. fornesi</i>	62	64	Brazil: DF, GO, PE	3, 9, 12
<i>O. fulvescens</i>	54	68	Costa Rica	6
<i>O. fulvescens</i>	60	74	Mexico	7
<i>O. longicaudatus</i> var.3	62	74,76	Venezuela	6
<i>O. longicaudatus</i> var.1	68	74,76	Peru	6
<i>O. microtis</i>	64	66	Peru	6
<i>O. nigripes</i>	62	80–82	Paraguay; Brazil: SP, RJ, ES, MG, SC, RS	4, 5, 9, 10, 13 14,
<i>O. nigripes</i>	62	78	Brazil: BA	10
<i>Oligoryzomys</i> sp.1	46	50	Brazil: BA	11
<i>Oligoryzomys</i> sp.2	44–45	52–53	Brazil: MG	11
<i>O. stramineus</i>	52	68–70	Brazil: GO, PE	3, 9
<i>Oligoryzomys</i> sp.	70	74,76	Brazil: GO,TO	This work

Acronyms for Brazilian states are: RS = Rio Grande do Sul, PR = Paraná, SP = São Paulo, RJ = Rio de Janeiro, ES = Espírito Santo, MG = Minas Gerais, GO = Goiás, DF = Distrito Federal, TO = Tocantins and PE = Pernambuco.

Cerrado, with a restricted distribution in the Rio Tocantins basin.

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REFERENCES

- Almeida, E. J. C. and Yonenaga-Yassuda, Y. 1991. Pericentric inversions and sex chromosome heteromorphisms in *Oryzomys nigripes* (Rodentia, Cricetidae). – *Caryologia* 44: 63–73.
- Aniskin, V. M. and Volobouev, V. T. 1999. Comparative chromosome banding of two South-American species of rice rats of the genus *Oligoryzomys* (Rodentia, Sigmodontinae). – *Chromosome Res.* 7: 557–562.
- Bonvicino, C. R. and Weksler, M. A. 1998. A new species of *Oligoryzomys* (Rodentia, Sigmodontinae) from northeastern and central Brazil. – *Z. Säugetierk.* 63: 90–103.
- Bonvicino, C. R., D'Andrea, P. S. and Borodin, P. 2001. Pericentric inversions: a study in natural populations of *Oligoryzomys nigripes* (Rodentia: Sigmodontinae). – *Genome* 44: 791–796.
- Brum-Zorrilla, G., Fronza, T. G., Wainberg, R. et al. 1988. *Oryzomys flavescens* and *O. delticola* chromosomes (Rodentia, Cricetidae) from Uruguay and Argentina. – *Caryologia* 41: 275–288.
- Espinosa, M. B. and Reig, A. O. 1991. Cytogenetics and karyosystematics of South American Oryzomyine rodents (Cricetidae, Sigmodontinae) III. Banding karyotypes of Argentinean *Oligoryzomys*. – *Z. Säugetierk.* 56: 306–317.
- Furtado, V. V. 1981. Diversidade cromossômica em roedores das famílias Cricetidae e Caviidae de Pernambuco, Brasil. – PhD thesis. Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Brazil.
- Gardner, A. L. and Patton, J. L. 1976. Karyotypic variation in Oryzomyine rodents (Cricetinae) with comments on chromosomal evolution in the neotropical cricetinae complex. – *Occas. Pap. Mus. Zool. L.A. State Univ.* 49: 1–47.
- Haiduk, M. W., Bickham, J. W. and Schimidly, D. J. 1979. Karyotypes of six species of *Oryzomys* from Mexico and Central America. – *J. Mammal.* 60: 610–615.

- 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.
- Musser, G. G. and Carleton, M. D. 1993. Family Muridae. – In: Wilson, D. E. and Reeder, D. M. (eds), *Mammal species of the world: a taxonomic and geographic reference*. Smithsonian Institution Press, pp. 501–755.
- Myers, P. and Carleton, M. D. 1981. The species of *Oryzomys* (*Oligoryzomys*) in Paraguay and the identity of Azara's "rat sixième ou rat à tarse noir". – *Misc. Publ. Mus. Zool. Univ. Mich.* 161: 1–14.
- Myers, P., Ludrigan, B. and Tucker, P. K. 1995. Molecular phylogenetics of Oryzomyine rodents: the genus *Oligoryzomys*. – *Mol. Phylogenet. Evol.* 4: 372–382.
- Sbalqueiro, I. J., Mattevi, M. S., Oliveira, L. F. B. et al. 1991. B chromosome system in populations of *Oryzomys flavescens* (Rodentia, Cricetidae) from Southern Brazil. – *Acta Theriol.* 36: 193–199.
- Seabright, M. 1971. A rapid banding technique for human chromosomes. – *Lancet* 2: 971–972.
- Silva, M. J. J. and Yonenaga-Yassuda, Y. 1997. New karyotypes of two related species of *Oligoryzomys* genus (Cricetidae, Rodentia) involving centric fusion with loss of NORs and distribution of telomeric (TTAGGG)_n sequences. – *Hereditas* 127: 217–229.
- Sumner, A. T. 1972. A simple technique for demonstrating centromeric heterochromatin. – *Exp. Cell Res.* 75: 304–306.
- Svartman, M. 1989. Levantamento cariotípico de roedores da região do Distrito Federal. – Master thesis, Instituto de Biociências, Universidade de São Paulo, Brazil.
- Yonenaga, Y., Frota-Pessoa, O., Kasahara, S. et al. 1976. Cytogenetic studies on Brazilian rodents. – *Ciênc. Cult.* 28: 202–211.
- Zanchin, N. I. T. 1988. Estudos cromossômicos em Oryzomys e Equimídeos da Mata Atlântica. – Master thesis. Porto Alegre, Universidade Federal do Rio Grande do Sul, Brazil.