Genetic Structure of Brazilian Populations of *Triatoma sordida* (Stål, 1859) (Hemiptera, Triatominae) by Means of Chromosomal Markers

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Abstract. *Triatoma sordida* is among the main Brazilian species considered as Chagas disease vectors. The genetic studies are directed mainly to phylogenetic questions because this species possibly have suffered cryptic speciation. Furthermore, there are few studies that analyzed the structure and genetic variability of specimens from Brazil and that showed low genetic diversity and strong genetic structuring of the population samples. Therefore, because of great epidemiological importance of *T. sordida* and mainly the restriction of genetic characterization of this vector only for populations of Minas Gerais state, this article performs a genetic analysis of the *T. sordida* from seven different Brazilian states (representing different biomes), by means of cytogenetic markers. All analyzed specimens presents the same cytogenetic characteristics: early meiotic prophase with several heterochromatic bodies dispersed in the nucleus (CG-rich), being one of them formed by the associated sex chromosomes surrounded by some autosomal heterochromatic regions, meiotic metaphase with most autosomal pairs exhibiting a C-heterochromatic block in one chromosomal end (CG-rich), Y sex chromosome fully heterochromatin (AT-rich), and X chromosome may present a small C-block (CG-rich). These results are important because the chromosomal markers enable to confirm and expand the low genetic diversity for all Brazilian states occupied by *T. sordida*, suggesting that all Brazilian populations were originated from a small ancestral population and possibly dispersed to other biomes by founder effect. In addition, we suggest that *T. sordida* from Brazil are not suffering cryptic speciation and we confirm the classification of all Brazilian examples as *T. sordida* sensu stricto.

INTRODUCTION

Chagas disease is a neglected disease considered as a potentially life-threatening illness caused by the protozoan *Trypanosoma cruzi* (Chagas, 1909), distributed mainly in endemic areas of 21 Latin American countries, where it is mostly vector-borne transmitted to humans by contact with feces of triatomines.¹ It is estimated that about eight million people are infected worldwide, mostly in Latin America where Chagas disease is endemic.¹

Presently, there are 154 species of triatomines, distributed in 18 genera and five tribes, being all species considered as potential vector of Chagas disease.^{2–4} In Brazil, there are 68 species distributed throughout the country's 27 states, and 64% of them are endemic species.^{5–8} *Triatoma infestans* (Klug, 1834), *Triatoma brasiliensis* (Neiva, 1911), *Triatoma pseudomaculata* (Corrêa and Espínola, 1964), *Triatoma sordida* (Stål, 1859), and *Panstrongylus megistus* (Burmeister, 1835) are the main species of triatomines related to the domestic transmission of Chagas disease.^{5,9}

Triatoma sordida is distributed in 14 Brazilian states (Acre, Bahia, Goiás, Mato Grosso, Mato Grosso do Sul, Maranhão, Minas Gerais, Paraná, Pernambuco, Piauí, Rio Grande do Sul, Santa Catarina, São Paulo, and Tocantins)^{5,10,11} and is considered the species with the highest frequency of domiciliary captures in Brazil.⁵ Although *T. sordida* exhibits endemism centered in the Cerrado, this species is also prevalent in Bahia

and the states of the Southeast and Midwest regions of Brazil, including transition areas to the Amazon,^{5,11} occupying thus other biomes besides the Cerrado: Pantanal, Atlantic forest, and Caatinga.¹⁰

The epidemiological importance of *T. sordida* is increasing because of its tendency to invade houses, particularly in areas where *T. infestans* has been controlled,^{12–14} being thus considered as “semi-domestic.”¹⁵ The anthropic activities have influence in the richness of this species because the areas of higher occurrence of *T. sordida* are the ones related to the agricultural activities in the past, which could explain its presence in areas that suffered ecologic impact due to significant loss of vegetation.¹⁶ Furthermore, *T. sordida* also is associated with the reinfestation of dwellings treated with insecticides.¹⁷

Although *T. sordida* exhibits wide geographic distribution in Brazil,^{5,10,11} genetic studies are directed mainly to phylogenetic questions^{18–22} because this species is also found in Argentina, Bolivia, Paraguay, and Uruguay⁵ and possibly has suffered cryptic speciation.^{19–21} Furthermore, there are few studies that analyzed the genetic structure and genetic variability of specimens from Minas Gerais, Brazil, that showed low genetic diversity and strong genetic structuring of the population samples.^{22,23}

Recently, Pessoa et al.²³ related low genetic diversity and strong genetic structuring of the population samples of *T. sordida* with five possible factors: possible geographical isolation due to an obstacle to Triatominae flow between neighboring localities; focal distribution of insects in small colonies usually comprised a few individuals, thereby limiting gene flow between them; the low dispersal capacity of *T. sordida*; the long Triatominae life cycle, which ensures that contributions from young adults able to reproduce (and consequently exchange genetic material) occurs over long

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intervals that differ among triatomine species; and continuous pressure from insecticides used since the 1950s to triatomines control.

As Chagas disease has no cure and treatment with benznidazole and nifurtimox are more effective in the acute phase of the disease (which is often asymptomatic),¹ vector control is the most effective method of preventing this neglected disease¹ and all knowledge about these hematophagous insects is important and can generate subsidies to assist the vector control programs. Thus, because of great epidemiological importance of *T. sordida* in Brazil and mainly the restriction of genetic characterization of this vector only for populations of Minas Gerais state, this article performs a genetic analysis of the *T. sordida* from seven different Brazilian states representing different biomes, by means of cytogenetic markers.

MATERIAL AND METHODS

Material. One hundred adult males coming from the municipalities described in Table 1 (at a minimum three copies of each locality) were used in the study (males were used because spermatogenesis is continued in adulthood, allowing the visualization of chromatin and chromosomes). The triatomines were provided by the insectariums of Faculdade de Ciências Farmacêuticas, Universidade Estadual Paulista "Júlio de Mesquita Filho", Araraquara, São Paulo, Brazil, of Instituto Oswaldo Cruz (IOC/FIOCRUZ), Rio de Janeiro, Rio de Janeiro, Brazil, and of Centro de Pesquisas René Rachou (CPqRR/FIOCRUZ), Belo Horizonte, Minas Gerais, Brazil. The characterization of biomes was performed according to the Brazilian Institute of Geography and Statistics.²⁴

Chromosome preparation. For cytological preparations, testes were removed from adult insects alive, fixed in an ethanol–acetic acid (3:1), and stored at –20°C. Squashes were made in a 50% acetic acid drop, coverslips were removed after freezing in liquid nitrogen, and the slides were air-dried and then stored at 4°C.

Chromomycin A3 (CMA3)/4,6-diamidino-2-phenylindole (DAPI) banding. C-banding was performed according to Sumner²⁵ for the characterization of constitutive heterochromatin

and CMA₃/DAPI banding was performed according to Schmid,²⁶ with modifications according to Severi-Aguiar et al.,²⁷ for differentiating the regions of heterochromatin rich in AT (DAPI⁺) and CG (CMA₃).

Analysis of biological material. At least 50 cells in prophase (for analysis of the chromatin) and 50 in metaphase (for analysis of the chromosomes) of *T. sordida* from each location were analyzed. The analysis of C-banding preparations was made using a Jenaval light microscope (Zeiss) attached to a digital camera and an Axio Vision LE 4.8 image analyzer (copyright 2006–2009 Carl Zeiss Imaging Solutions GmbH) and of CMA₃/DAPI banding preparation was analyzed using a fluorescence microscopy Zeiss-Axioskop and Olympus BX-FLA.

RESULTS

All analyzed specimens, regardless the locality (Table 1), present the same cytogenetic characteristic: C-banding: during early meiotic prophase, several heterochromatic bodies are observed, being one of them formed by the associated sex chromosomes surrounded by some autosomal heterochromatic regions (Figure 1A, *); Furthermore, during the meiotic metaphase, the most autosomal pairs present a C-heterochromatic block in one chromosomal end and X chromosome may present a small C-block (Figure 1B) (Y sex chromosome is heterochromatin in all species of triatomine); CMA₃/DAPI banding: several bodies CG-rich dispersed in the nucleus of early meiotic prophase, being one of them formed by the association of the X sex chromosome surrounded by some autosomal (Figure 1C, *) and Y sex chromosome AT-rich DNA (Figure 1D).

DISCUSSION

Cytogenetic markers have been used because initially Panzera et al.¹⁹ differentiated *T. sordida* from Brazil and Argentina by the pattern of heterochromatin. Recently, Panzera et al.²¹ and Bardella et al.²⁸ recognized three chromosomal taxa for *T. sordida* (*T. sordida sensu stricto*, *T. sordida Argentina* and *T. sordida La Paz*). Panzera et al.²¹ analyzed Brazilian representatives from Minas Gerais, Piauí, and Mato Grosso with C-banding and observed the same results that we observed for all analyzed specimens. Beyond that, we characterized the composition of DNA rich in AT and CG in chromatin of these vectors, and all the specimens of *T. sordida* analyzed showed the same disposition in chromatin and chromosomes.

These results are important because the chromosomal markers enable confirming and expanding the results described by Pessoa et al.²³ and Monteiro et al.²² for all Brazilian states occupied by *T. sordida*. In addition, the low genetic diversity can also be confirmed by the low genetic distance observed between *T. sordida* from different Brazilian states [e.g., 0.011 among populations of Mato Grosso (Pocotó) and Mato Grosso do Sul (Pantanal) with the cytochrome oxidase I gene; 0.007 among populations of Mato Grosso (Rondonópolis) and Mato Grosso do Sul (Pantanal) with cytochrome oxidase II gene; and 0.006 among populations from municipalities in the state of Mato Grosso (Rondonópolis and Pocotó), and 0.004 and 0.002 distance observed between Rondonópolis and Pantanal and Pocotó and Pantanal,

TABLE 1

Geographical origin of the specimens used in the study and their corresponding biomes

<i>Triatoma sordida</i> collection sites		
Brazilian states	Municipalities	Brazilian biomes
Mato Grosso	Rondonópolis	Cerrado
Goiás	Posse	Cerrado
	São Luiz dos Montes	Cerrado
Minas Gerais	Belos	
	Matias Cardoso and Monte Azul	Cerrado and Caatinga
Bahia	Itinga	Mata Atlântica
	Seabra and Macaúbas	Caatinga
Rio Grande do Sul	São Francisco de Assis	Pampa and Mata Atlântica
São Paulo	Mirassol and Guairá	Mata Atlântica and Cerrado
	São Paulo	Mata Atlântica
Mato Grosso do Sul	Rio Verde de Mato Grosso	Cerrado and Pantanal
	Corumbá	Pantanal
	Brasilândia	Mata Atlântica and Cerrado

FIGURE 1. Constitutive heterochromatin pattern and composition of AT-rich and CG-rich DNA observed in *Triatoma sordida* Brazilians. (A) Early meiotic prophase prepared with C-banding. Note the several heterochromatic bodies dispersed in the nucleus, being one of them formed by the associated sex chromosomes surrounded by some autosomal heterochromatic regions (*). (B) Meiotic metaphase prepared with C-banding. Note that the most autosomal pairs present a C-heterochromatic block in one chromosomal end, Y sex chromosome is fully heterochromatic and X chromosome may present a small C-block. (C and D) Early meiotic prophase prepared with CMA₃/DAPI staining. (C) Note the several bodies CG-rich dispersed in the nucleus, being one of them formed by the associated X sex chromosome surrounded by some autosomal (*). (D) Note that only the Y sex chromosome is AT-rich. X: X sex chromosome Y: Y sex chromosome. Bar = 10 µm. This figure appears in color at www.ajtmh.org.

respectively, both with the 16S r DNA gene²⁹ and by the low variability of the wing geometry of *T. sordida* observed recently among populations from Brazil.³⁰

Some authors suggest that *T. sordida* probably have originated in the Brazilian Cerrado^{16,18,31} and as a consequence of deforestation and *T. infestans* control strategies, it has dispersed toward the south.^{13,14,32} Our results also suggest that all Brazilian populations originated from a small ancestral population (possibly in the Cerrado, as suggested previously) and dispersed to other biomes by the founder effect (possibly by active dispersal by flight³³ or passive dispersion by nymphs associated with bird feathers³⁴). However, because there are evidences that genetic diversity of *T. infestans* is reduced in areas treated with insecticides because of bottleneck events,^{35,36} we cannot also dismiss this hypothesis for *T. sordida*.

Noireau et al.²⁰ reported for the first time the presence of cryptic speciation to *T. sordida* specimens from Bolivia. Monteiro et al.²² analyzed this phenomenon for four populations from Minas Gerais, Brazil, and found no cryptic speciation. Our results demonstrate that *T. sordida* Brazilians are not suffering cryptic speciation, because if this evolutionary event was taking place, the results with chromosomal markers would demonstrate differences, and were recently observed for specimens of Bolivia.²¹

Besides its importance for the genetic and evolutionary characterization of Brazilian *T. sordida*, cytogenetic analysis

confirms that all triatomine present in Brazilian insectariums, initially classified as *T. sordida* by morphology, exhibit the same chromosome pattern characterized by Panzera et al.²¹ as *T. sordida* sensu stricto, assisting directly in the taxonomy of these vectors. In addition, for the first time, *T. sordida* specimens from several Brazilian biomes and mainly representing areas of peridomicile (Rondonópolis and Guaira chicken coops), intradomicile (Mirassol and Miranda's houses), and wild (Corumbá, associated with bird nests) had their genetic diversity analyzed. The low genetic variability observed for *T. sordida* can be an important tool for the control of this species by the vector programs.

Thus, by means of the cytogenetic study of Brazilian *T. sordida*, it was observed that this vector species, although widely distributed in Brazil, has extremely low genetic diversity. Moreover, we confirm the classification of all Brazilian examples as *T. sordida* sensu stricto. Our results are important to direct further studies (such as phylogeography) and, especially, to assist in developing tools for the control of this vector species by vector control programs.

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REFERENCES

1. World Health Organization, 2018. *Chagas Disease (American Trypanosomiasis)*. Available at: <http://www.who.int/chagas/en/>. Accessed April 18, 2018.
2. Oliveira J, Alevi KCC, 2017. Taxonomic status of *Panstrongylus herzeri* Wygodzinsky, 1948 and the number of chagas disease vectors. *Rev Soc Bras Med Trop* 50: 434–435.
3. Oliveira J, Ayala JM, Justi SA, Rosa JA, Galvão C, 2018. Description of a new species of *Nesotriatoma* Usinger, 1944 from Cuba and revalidation of synonymy between *Nesotriatoma bruneri* (Usinger, 1944) and *N. flavida* (Neiva, 1911) (Hemiptera, Reduviidae, Triatominae). *J Vector Ecol* 43: 148–157.
4. Dom PL, Just SA, Dale C, Stevens L, Galvão C, Cordon RL, Monroy C, 2018. Description of *Triatoma mopan* sp. n. (Hemiptera, Reduviidae, Triatominae) from a cave in Belize. *Zookeys* 775: 69–95.
5. Galvão C, 2014. *Vetores da doença de chagas no Brasil*. Curitiba, Brazil: Sociedade Brasileira de Zoologia.
6. Mendonça VJ, Alevi KCC, Pinotti H, Gurgel-Gongalves R, Pita S, Guerra AL, Panzera F, Araújo RF, Azeredo-Oliveira MTV, Rosa

- JA, 2016. Revalidation of *Triatoma bahiensis* Sherlock & Serafim, 1967 (Hemiptera: Reduviidae) and phylogeny of the *T. brasiliensis* species complex. *Zootaxa* 4107: 239–254.
7. Souza ES, Atzinger NCBV, Furtado MB, Oliveira J, Damieli JN, Vendramini DP, Gardim S, Rosa JA, 2016. Description of *Rhodnius marabaensis* sp. N. (Hemiptera: Reduviidae: Triatominae) from Pará State, Brazil. *Zookeys* 621: 45–62.
 8. Rosa JA, Justino HHG, Nascimento JD, Mendonça VJ, Rocha CS, Carvalho DB, Falcone R, Azeredo-Oliveira MTV, Alevi KCC, Oliveira J, 2017. A new species of *Rhodnius* from Brazil (Hemiptera, Reduviidae, Triatominae). *ZooKeys* 675: 1–25.
 9. Silveira AC, Rezende DF, 1984. Epidemiologia e controle da transmissão vetorial da doença de Chagas no Brasil. *Rev Soc Bras Med Trop* 27: 11–22.
 10. Gurgel-Gonçalves R, Galvão C, Costa J, Peterson AT, 2012. Geographic distribution of chagas disease vectors in Brasil based on ecological niche modeling. *J Trop Med* 2012: 1–15.
 11. Ramos LJ, Souza JL, Souza CR, Oliveira J, Rosa JA, Camargo LMA, Cunha RM, Meneguetti DUO, 2018. First report of *Triatoma sordida* Stål, 1859 (Hemiptera, Reduviidae, Triatominae) in the state of Acre and Brazilian western Amazon. *Rev Soc Bras Med Trop* 51: 77–79.
 12. Forattini OW, Rabello EX, Ferreira OA, Rocha e Silva EO, Santos JLF, 1984. Aspectos ecológicos da tripanossomíase americana. XXI. Comportamento de espécies triatomíneas silvestres na reinfestação do intra e peridomicílio. *Rev Saude Publica* 18: 185–208.
 13. Diotaiuti L, 1995. Potencial vetorial do *Triatoma sordida*. *Rev Soc Bras Med Trop* 28: 38–41.
 14. Diotaiuti L, de Paula OR, Falcão PÇ, Pinto Dias JC, 1995. Avaliação de programa de controle vetorial da doença de chagas em Minas Gerais, Brasil, com referencia especial ao *Triatoma sordida*. *Bol Ojicina Sanit Panam* 118: 11–219.
 15. Coura JR, 1993. O falso dilema sobre a luta antivetorial e as perspectivas de controle da doença de chagas no Brasil. *Cad Saude Publica* 9: 514–518.
 16. Forattini OP, 1980. Biogeography, origin and distribution of Triatominae domicile dispersal in Brazil. *Rev Saude Publica* 14: 265–299.
 17. Pessoa GCD, Obara MT, Rezende JC, de Mello BV, Ferraz ML, Diotaiuti L, 2015. Deltamethrin toxicological profile of peridomestic *Triatoma sordida* in the north of Minas Gerais, Brazil. *Paras Vect* 8: 263.
 18. Gorla DE, Jurberg J, Catala S, Schofield CJ, 1993. Systematics of *Triatoma sordida*, *T. guasayana* and *T. patagonica* (Hemiptera—Reduviidae). *Mem Inst Oswaldo Cruz* 88: 379–385.
 19. Panzera F, Hornos S, Pereira J, Cestau R, Canale D, Diotaiuti L, Dujardin JP, Prrez R, 1997. Genetic variability and geographic differentiation among three species of triatomine bugs (Hemiptera: Reduviidae). *Am J Trop Med Hyg* 57: 732–739.
 20. Noireau F, Gutierrez T, Zegarra M, Flores R, Brenière F, Cardozo L, Dujardin JP, 1998. Cryptic speciation in *Triatoma sordida* (Hemiptera: Reduviidae) from the Bolivian Chaco. *Trop Med Int Health* 3: 364–372.
 21. Panzera F, Pita S, Nattero J, Panzera Y, Galvão C, Chavez T, Rojas de Arias A, Cardozo Tellez L, Noireau F, 2015. Cryptic speciation in the *Triatoma sordida* subcomplex (Hemiptera, Reduviidae) revealed by chromosomal markers. *Paras Vect* 8: 495–504.
 22. Monteiro FA, Jurberg J, Lazoski C, 2009. Very low levels of genetic variation in natural peridomestic populations of the chagas disease vector *Triatoma sordida* (Hemiptera: Reduviidae) in southeastern Brazil. *Am J Trop Med Hyg* 81: 223–227.
 23. Pessoa GCA, Sousa TN, Sonoda IV, Diotaiuti L, 2016. Assessing the mitochondrial DNA diversity of the chagas disease vector *Triatoma sordida* (Hemiptera: Reduviidae). *Mem Inst Oswaldo Cruz* 111: 322–329.
 24. Instituto Brasileiro de Geografia e Estatística—IBGE, 2016. *Cidades*. Available at: <https://cidades.ibge.gov.br/>. Accessed April 18, 2018.
 25. Sumner AT, 1972. A simple technique for demonstrating centromeric heterochromatin. *Exp Cell Res* 75: 305–306.
 26. Schimid M, 1980. Chromosoma banding an amphibia IV. Differentiation of GC and AT rich regions in Anura. *Chromosoma* 77: 83–103.
 27. Severi-Aguiar GD, Lourenço LB, Bicudo HE, Azeredo-Oliveira MTV, 2006. Meiosis aspects and nucleolar activity in *Triatoma vitticeps* (Triatominae, Heteroptera). *Genetica* 126: 141–151.
 28. Bardella VB, Pita S, Vanzela ALL, Galvão C, Panzera F, 2016. Heterochromatin base pair composition and diversification in holocentric chromosomes of kissing bugs (Hemiptera, Reduviidae). *Mem Inst Oswaldo Cruz* 111: 614–624.
 29. Justi SA, Russo CAM, Mallet JRS, Obara MT, Galvão C, 2014. Molecular phylogeny of Triatomini (Hemiptera: Reduviidae: Triatominae). *Paras Vect* 7: 149.
 30. Vendramini DP, Obara MT, Gurgel-Gonçalves R, Ceretti-Junior W, Marrelli MT, 2017. Wing geometry of *Triatoma sordida* (Hemiptera: Reduviidae) populations from Brazil. *Infect Genet Evol* 49: 17–20.
 31. Forattini OP, Barata JMS, Santos JLF, Silveira AC, 1982. Hábitos alimentares, infecção natural e distribuição de triatomíneos domiciliados na região central do Brasil. *Rev Saude Pública* 16: 171–204.
 32. Dias JCP, 1988. Controle de vetores da doença de chagas no Brasil e risco de reinvasão domiciliar por vetores secundários. *Mem Inst Oswaldo Cruz* 83: 387–397.
 33. Schofield CJ, Lehane MJ, McEwan P, Catalá SS, Gorla DE, 1991. Dispersive flight by *Triatoma sordida*. *Trans R Soc Trop Med Hyg* 85: 676–678.
 34. Forattini OP, Rocha e Silva EO, Ferreira OA, Rabello EX, Pattoli DG, 1971. Ecological aspects of trypanosomiasis. 3. Local dispersion of Triatominae with special reference to *Triatoma sordida*. *Rev Saude Publica* 5: 193–205.
 35. Pérez de Rosas AR, Segura EL, Fichera L, Garcia BA, 2008. Macrogeographic and microgeographic genetic structure of the chagas disease vector *Triatoma infestans* (Hemiptera: Reduviidae) from Catamarca, Argentina. *Genetica* 133: 247–260.
 36. Pérez de Rosas AR, Segura EL, García BA, 2007. Microsatellite analysis of genetic structure in natural *Triatoma infestans* (Hemiptera: Reduviidae) populations from Argentina: its implication in assessing the effectiveness of chagas disease vector control programmes. *Mol Ecol* 16: 1401–1412.