



Article/Artigo

Occurrence of positivity for *Trypanosoma cruzi* in triatomine from municipalities in Southeastern Brazil, from 2002 to 2004

Ocorrência e positividade para *Trypanosoma cruzi* em triatomíneos de municípios da região sudeste do Brasil, de 2002 a 2004

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ABSTRACT

Introduction: from an epidemiological point of view, more than 120 species of triatomine (Hemiptera, Reduviidae) are known. The occurrence and positivity for *Trypanosoma cruzi* in triatomines in 16 municipalities of the Triângulo Mineiro and Alto Paranaíba were evaluated from January 2002 to December 2004. **Methods:** the triatomines were captured basically according to the classic norms of the National Health Foundation. The parasitological exams of the triatomines were conducted according to the technique described by the Ministry of Health. During the study period, 990 specimens of triatomines were captured and of these, 771 could be examined. **Results:** five species were identified: *Triatoma sordida*, *Panstrongylus diasi*, *Panstrongylus megistus*, *Panstrongylus geniculatus* and *Rhodnius neglectus*. *Triatoma sordida* represented 71.5% of all the triatomines captured, followed by *Panstrongylus megistus* (18%), *Rhodnius neglectus* (9.3%), *Panstrongylus diasi* (0.8%) and *Panstrongylus geniculatus* (0.4%). Of the total number of triatomines examined, 2.7% were positive for *Trypanosoma cruzi*. *Panstrongylus megistus* was the species that presented the highest rates of infection by *Trypanosoma cruzi* (8.3%), followed by *Rhodnius neglectus* (2.9%) and *Triatoma sordida* (1.4%). **Conclusions:** there is a need to adapt to new circumstances in epidemiology, with greater emphasis on entomological surveillance, since the potential for adaptation of secondary species of triatomines exists, especially where Chagas' disease is already under control.

Key-words: Triatomine. Chagas' disease. Epidemiology. *Trypanosoma cruzi*. Brazil.

RESUMO

Introdução: do ponto de vista epidemiológico mais de 120 espécies de triatomíneos (Hemiptera, Reduviidae) são conhecidas. A ocorrência e a positividade de *Trypanosoma cruzi* em triatomíneos de 16 municípios do Triângulo Mineiro e Alto Paranaíba foram avaliadas de janeiro de 2002 a dezembro de 2004. **Métodos:** os triatomíneos foram capturados seguindo basicamente as normas clássicas da Fundação Nacional de Saúde. Os exames parasitológicos dos triatomíneos foram conduzidos de acordo com a técnica descrita pelo Ministério da Saúde. Durante o período de estudo, foram capturados 990 exemplares de triatomíneos, sendo que 771 dos capturados estavam em condições de serem examinados. **Resultados:** cinco espécies foram identificadas: *Triatoma sordida*, *Panstrongylus diasi*, *Panstrongylus megistus*, *Panstrongylus geniculatus* e *Rhodnius neglectus*. *Triatoma sordida* representou 71,5% de todos os triatomíneos capturados, seguido por *Panstrongylus megistus* (18%), *Rhodnius neglectus* (9,3%), *Panstrongylus diasi* (0,8%) e *Panstrongylus geniculatus* (0,4%). Dos triatomíneos examinados, 2,7% foram positivos para *Trypanosoma cruzi*. *Panstrongylus megistus* foi a espécie que apresentou a maior taxa de infecção por *Trypanosoma cruzi* (8,3%), seguida pelo *Rhodnius neglectus* (2,9%) e *Triatoma sordida* (1,4%). **Conclusões:** há necessidade de se adequar às novas circunstâncias epidemiológicas com ênfase na vigilância entomológica, uma vez que o potencial de adaptação de espécies secundárias de triatomíneos, em áreas onde a doença de Chagas está controlada, é uma preocupação.

Palavras-chaves: Triatomíneos. Doença de Chagas. Epidemiologia. *Trypanosoma cruzi*. Brasil.

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Received in 20/04/2009

Accepted in 16/12/2009

INTRODUCTION

From an epidemiological point of view, more than 120 species of triatomine (Hemiptera, Reduviidae) have already been identified. They are strictly hematophagous insects, generally with nocturnal habits and many are potential vectors of Chagas' disease (CD), an anthroponosis known as American trypanosomiasis¹⁻³.

In Brazil, the control of CD vectorial transmission was institutionalized by the National Malaria Service in 1950. An entomological survey, conducted between 1975 and 1983, demonstrated that of more than 42 species of triatomines identified in Brazil, 30 were detected in the domestic environment. Five of these were considered to be active in the transmission of Chagas: *Triatoma infestans*, *Panstrongylus megistus*, *Triatoma brasiliensis*, *Triatoma pseudomaculata* and *Triatoma sordida*⁴. Satisfactory results were obtained with the consolidation of control programs in relation to the domestic environment. The *Triatoma infestans* infestation was controlled and entomological surveillance of the secondary triatomine species was established⁵⁻⁸. In spite of the magnitude of the problems represented by CD and the proven efficacy of insecticides with residual action in the combat of the vector, control was only systematized and structured in a nationwide program in the 1970s⁹. In July 2006, the elimination of vectorial transmission of CD by *Triatoma infestans* was certified in all states of Brazil. This process involved various actions, including the sanitization and inspection of residences. The result was a strong decrease in the number of children infected, as well as 100% control of contamination in blood banks as prevalence of the disease was reduced to 0.6% of the donors¹⁰. However other species of triatomine that are normally of secondary importance in the transmission of *Trypanosoma cruzi* in humans, have been captured with increasing frequency, and, surprisingly, in high densities within dwellings, due to the growing environmental changes caused by humans.

Since 2000, the control of CD in the State of Minas Gerais occurs in a decentralized manner, as the municipalities assume the responsibility for implementing such activities. The responsibility for monitoring triatomines is the municipalities, which perform scheduled visits in homes. The municipality performs this function through visitations by a health worker who searches the residence and sprays when the presence of triatomines is confirmed¹¹. The State of Minas Gerais received certification for eliminating CD through transmission by *Triatoma infestans* from the Pan American Health Organization in 2002. In addition, from 1989 to 2006, 34,000 samples of human sera originating from 408 municipalities were tested and less than 1% of samples presented positive results, demonstrating the absence of recent infection caused by triatomine bites. Most of the deaths, about 99.88%, occurred in patients with chronic disease, aged 20 years-old or over, which means they are remnants of the period in which vectorial transmission had not yet been interrupted¹¹.

The aim of this study was to evaluate the occurrence and positivity for *Trypanosoma cruzi* in triatomines in 16 municipalities of the Triângulo Mineiro and Alto Paranaíba region of Southeastern Brazil, from January, 2002 to December, 2004.

METHODS

Study area

The study area involved 16 municipalities in the Triângulo Mineiro and Alto Paranaíba region of the State of Minas Gerais, in Southeastern Brazil (**Figure 1**). Two of the 18 municipalities of the region were eliminated due to the nonexistence of data during the period. The region is part of the savanna or *cerrado* ecosystem. It is an area that has suffered great environmental impact during the last few decades due to unplanned urban growth. The native *cerrado* vegetation has been mostly substituted by cattle and

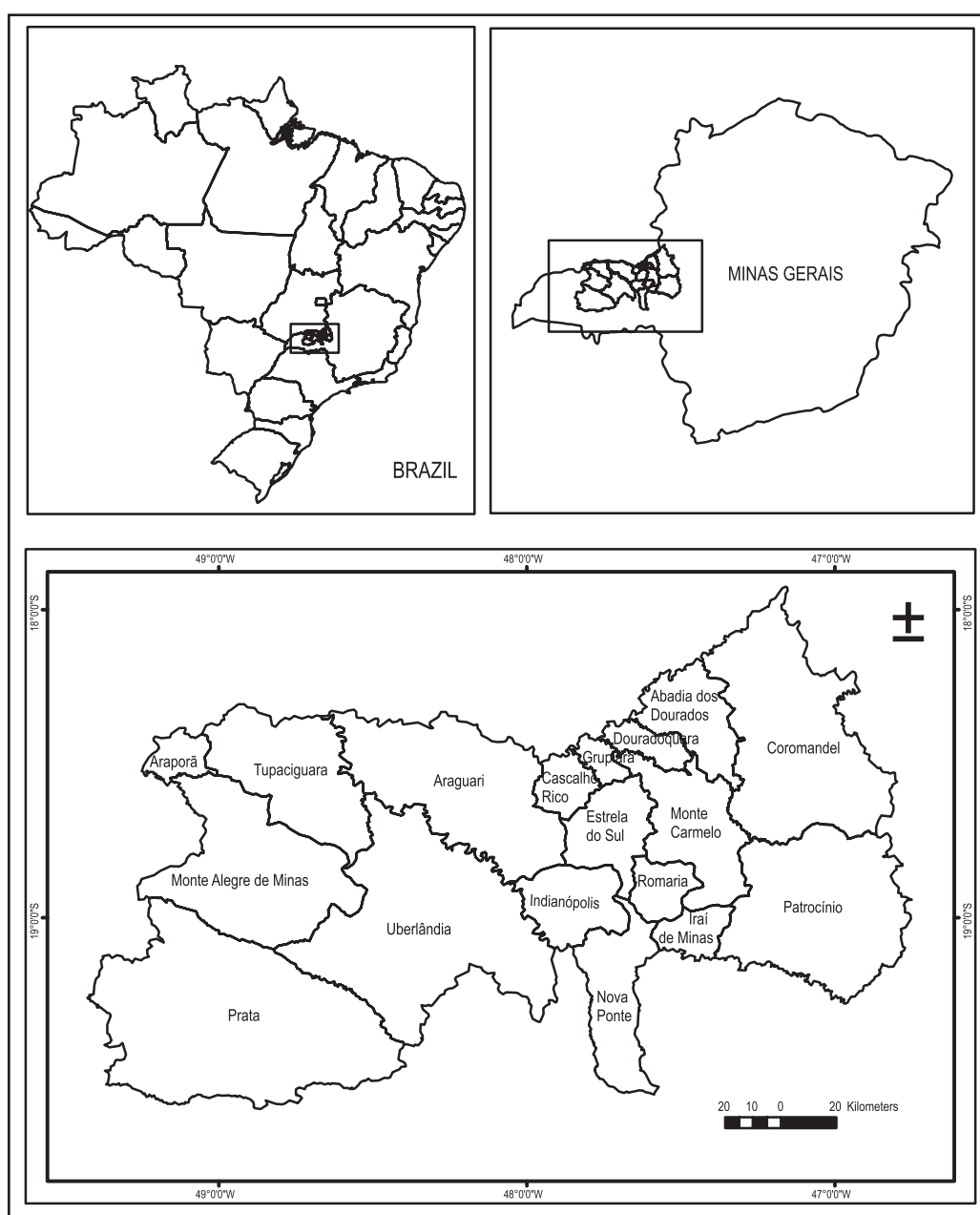


FIGURE 1 - Area covered by the Regional Health Administration of Uberlândia-Minas Gerais, Brazil.

crops, corn and soybeans, which have created extensive ecological imbalance.

The data referring to 16 municipalities were obtained from the Epidemiological Information Center in Uberlândia. This center is part of the State Health Department (SHD). Of the municipalities studied, only seven provided integral surveillance and systematic combat of triatomine. The governmental control programs were less effective in the other municipalities.

The triatomines were captured basically according to the classic norms of the National Health Foundation (*Fundação Nacional de Saúde*, FUNASA)¹² and refined by the Technical Management in CD SHD-MG. The triatomines were captured in three ways: 1) Reporting bugs: a service performed by the population, involving handing the suspected insect in to a Triatominae Information Center (TIC) installed at strategic locations in the municipalities, such as schools and health centers; 2) Notification service: performed by staff working in the Chagas' Disease Control Program (ChDCP) for each municipality, 3) Active searches: the search for vectors was conducted in households in the municipality, regardless of whether the population had performed any searches or registered any complaints. The households consisted of the house and annexes and other buildings (chicken run, rubbish dump, etc). The residents were advised by staff of the ChDCP concerning the likely places to find the insects and instructed on how to capture them with due care and send them to the nearest TIC.

Parasitological exams

After external morphological analysis of the triatomines, the specimens were identified according to growth stage, sex, genus and species. The insects that presented intestinal content were forwarded for feces examination to test for the presence of *Trypanosoma cruzi*. The parasitological exams of the triatomines were conducted according to the technique described by the Brazilian Ministry of Health¹³. The last abdominal segments of each triatomine were gently squeezed with tweezers and the fecal matter collected on a 25 by

75mm-diameter microscope slide. The material was homogenized in an isotonic solution of 0.9% NaCl, thin smears were obtained and colored with methylene blue for 1 sec. They were then washed with buffered water to remove excess stain. Subsequently, the smear was covered with freshly prepared Giemsa solution (7 drops of Giemsa/5ml solution in buffered water), for 30 min. After this period the slides were stored at room temperature in an inclined position, dried and the excessive coloring removed, until the moment of microscopic examination of the material, using 100X objectives and immersed in oil.

Statistical analysis

The Student t test was applied to verify any numerical differences for captured triatomines between the municipalities with systematic epidemiological surveillance and those that did not have such programs. Municipal comparisons were elaborated using the Mann-Whitney nonparametric test. The Binomial nonparametric test was used to compare proportions of positive exams between the municipalities evaluated¹⁴. The significance level was established at $p < 0.05$.

RESULTS

During the period studied, 990 specimens of triatomines were captured. The intestinal content of 771 specimens was examined, representing 78% of the total. The remaining specimens were dry and did not present intestinal contents (**Table 1**). Triatomines positive for *Trypanosoma cruzi* were detected in 50% of the 16 municipalities studied.

The municipalities that conducted systematic control activity were 48% of the regional total. The majority 762 (77%) of triatomines captured came from these municipalities. A significant difference ($p = 0.0017$) was observed between the number of triatomines captured in the cooperating municipalities and those that did not have systematic control. A significant difference ($p = 0.0027$) was

TABLE 1 - Numbers of triatomine captured, identified and examined as positive for *Trypanosoma cruzi* in 16 municipalities of the Uberlândia, Minas Gerais Regional Health Administration, during the period between January 2002 and December 2004.

Municipalities	2002			2003			2004			Total		
	NC	NE	Pos	NC	NE	Pos	NC	NE	Pos	NC	NE	Pos
Araporã*	0	0	0	11	12	0	182	148	2	193	160	2
Patrocínio*	75	64	8	22	8	0	21	15	0	120	87	8
Tupaciguara*	13	13	0	7	2	0	90	73	1	110	88	1
Araguari*	19	14	0	55	43	1	30	30	0	104	87	1
Douradoquara	25	19	0	41	28	2	34	24	1	100	71	3
Uberlândia*	14	12	1	37	25	1	48	46	2	99	83	4
Prata*	0	0	0	41	37	0	53	43	1	94	80	1
Monte Carmelo*	6	6	0	6	3	0	30	18	0	42	27	0
Abadia dos Dourados	19	10	0	5	4	0	9	4	0	33	18	0
Indianópolis	22	19	0	4	3	0	4	4	0	29	26	0
Coromandel	2	1	0	13	3	0	5	3	0	20	7	0
Grupiara	5	5	1	12	11	0	3	0	0	20	16	1
Estrela do Sul	0	0	0	8	6	0	5	4	0	13	10	0
Irai de Minas	0	0	0	6	5	0	3	2	0	9	7	0
Nova Ponte	0	0	0	2	2	0	1	1	0	3	3	0
Cascalho Rico	0	0	0	1	1	0	0	0	0	1	1	0
Total	200	163	10	271	193	4	518	415	7	990	771	21

*Municipalities that conducted systematic surveillance action and combat of triatomine in the Chagas' Disease Control Program.

NC: Number of triatomine captured; NE: Number of triatomine examined; Pos: Positive

also verified between the number of triatomines examined from cooperating municipalities and those without systematic control. The relation between the number of positive and examined triatomines, in the cooperating municipalities and those without systematic control, was compared, with no significant differences.

Five species were identified: *Triatoma sordida*, *Panstrongylus diasi*, *Panstrongylus megistus*, *Panstrongylus geniculatus* and *Rhodnius neglectus* (Table 2). There was a predominance of *Triatoma sordida* (71.5%) of the captured specimens, followed by *Panstrongylus megistus* (18%), *Rhodnius neglectus* (9.3%), *Panstrongylus diasi* (0.8%) and *Panstrongylus geniculatus* (0.4%). Of the total number of insects captured, 684 (69.1%) specimens came from the peridomicile area and 306 (30.9%) from the dwelling itself. *Triatoma sordida* was the species most frequently captured in the peridomicile area (82.2%) and in the intradomicile area (47.7%).

Of the total number of triatomines examined, 2.7% were positive for *Trypanosoma cruzi*. Among the insects captured in the intradomicile area, 3.4% of the positive specimens for *Trypanosoma cruzi* were *Rhodnius neglectus*, 2.9% were *Triatoma sordida* and 2.3% were *Panstrongylus megistus*. The species that presented the highest rate of infection by *Trypanosoma cruzi* was *Panstrongylus megistus* (8.3%), followed by *Rhodnius neglectus* (2.9%) and *Triatoma sordida* (1.4%). The other species examined did not present *Trypanosoma cruzi* infection.

Table 3 shows the identification of the triatomines captured, examined and those positive for *Trypanosoma cruzi*, distributed throughout the area studied. Specimens of *Panstrongylus megistus* and *Triatoma sordida* were captured in 14 (87.5%) of the 16 municipalities that conducted entomological surveillance. Specimens of *Rhodnius neglectus* were captured in 13 (81.3%) of these municipalities.

TABLE 2 - Localization of the five triatomine species captured, examined and positive for *Trypanosoma cruzi* in 16 municipalities of the Uberlândia, Minas Gerais Regional Health Administration, during the period between January, 2002 and December, 2004.

Genus/specie	Peridomiciliar					Intradomiciliar					Total				
	captured		examined			captured		examined			captured		examined		
	n	%	pos	n	%	n	%	pos	n	%	n	%	pos	n	%
<i>Triatoma sordida</i>	562	82.2	5	461	1.1	146	47.7	3	102	2.9	708	71.5	8	563	1.4
<i>Panstrongylus diasi</i>	1	0.1	0	1	0	7	2.3	0	5	0	8	0.8	0	6	0
<i>Panstrongylus megistus</i>	107	15.7	10	90	11.1	71	23.2	1	43	2.3	178	18	11	133	8.3
<i>Panstrongylus geniculatus</i>	1	0.1	0	0	0	3	1.0	0	1	0	4	0.4	0	1	0
<i>Rhodnius neglectus</i>	13	1.9	0	10	0	79	25.8	2	58	3.4	92	9.3	2	68	2.9
Total	684	69.1	15	562	2.7	306	30.9	6	209	2.9	990	100	21	771	2.7

n: number of triatomine captured or examined; pos: positive

TABLE 3 - Genus and species of triatomine captured, examined and positive for *Trypanosoma cruzi* in 16 municipalities covered by the Uberlândia-Minas Gerais Regional Health Administration during the period between January 2002 to December 2004.

Municipalities	<i>Triatoma sordida</i>			<i>Panstrongylus diasi</i>			<i>Panstrongylus megistus</i>			<i>Panstrongylus geniculatus</i>			<i>Rhodnius neglectus</i>			Total		
	NC	NE	pos	NC	NE	pos	NC	NE	pos	NC	NE	pos	NC	NE	pos	NC	NE	pos
Araporã*	191	158	2	2	2	0	-	-	-	-	-	-	-	-	-	193	160	2
Patrocínio*	4	3	0	1	1	0	104	76	8	-	-	-	11	7	0	120	87	8
Tupaciguara*	103	82	1	-	-	-	3	3	0	-	-	-	4	3	0	110	88	1
Araguari*	92	79	1	-	-	-	7	7	0	2	0	0	3	1	0	104	87	1
Douradoquara	70	46	2	-	-	-	5	4	1	1	0	0	24	21	0	100	71	3
Uberlândia*	71	59	2	5	3	0	7	7	1	-	-	-	16	14	1	99	83	4
Prata*	81	68	0	-	-	-	11	10	1	-	-	-	2	2	0	94	80	1
Monte Carmelo*	28	17	0	-	-	-	11	7	0	-	-	-	3	3	0	42	27	0
Abadia dos Dourados	15	7	0	-	-	-	2	2	0	-	-	-	16	9	0	33	18	0
Indianópolis	24	22	0	-	-	-	4	3	0	-	-	-	1	1	0	29	26	0
Coromandel	-	-	-	-	-	-	14	5	0	1	1	0	5	1	0	20	7	0
Grupiara	15	12	0	-	-	-	1	1	0	-	-	-	4	3	1	20	16	1
Estrela do Sul	12	9	0	-	-	-	1	1	0	-	-	-	-	-	-	13	10	0
Irai de Minas	1	0	0	-	-	-	7	6	0	-	-	-	1	1	0	9	7	0
Nova Ponte	-	-	-	-	-	-	1	1	0	-	-	-	2	2	0	3	3	0
Cascalho Rico	1	1	0	-	-	-	-	-	-	-	-	-	-	-	-	1	1	0
Total	708	563	8	8	6	0	178	133	11	4	1	0	92	68	2	990	771	21

*Municipalities that conducted systematic surveillance action to combat triatomine in the Chagas' Disease Control Program. NC: number of triatomine captured. NE: number of triatomine examined.

NC: number of triatomine captured, NE: number of triatomine examined, pos: number of triatomine examined and identified as positive for *Trypanosoma cruzi*.

DISCUSSION

Chagas' disease became a zoonotic disease when the traditional vectors, native to natural areas, were dislocated from their original wild ecotypes to human surroundings. The presence of the triatomines in dwellings occurred due to land use changes, such as clearing for crops, which has become prevalent in the region. Economic distortions and social differences also produce consequences that promote *Trypanosoma cruzi* transmission. In most areas where the insects have become prevalent, dwellings are precarious, incomes are obviously very low and social conditions are lamentable^{1,15}.

The persistence of residual foci of *Triatoma infestans* in some Brazilian states, as well as the existence of a large number of autochthonous species that are potential vectors commonly found within dwellings (*Panstrongylus megistus*, *Triatoma brasiliensis*, *Triatoma pseudomaculata* and *Triatoma sordida*), represent risk factors for CD transmission. In addition, sporadic cases of oral transmission have been reported¹⁶⁻¹⁸.

In the State of Minas Gerais, of the 853 municipalities, 345 (40.4%) are under epidemiological surveillance. However, in the year 2008, 517 municipalities were surveyed by the ChDCP. Considering this number, 66.7% of the municipalities conduct disease surveillance (ML Ferraz: Unpublished Data). Our findings permit the inference that municipalities with systematic control programs demonstrate proven efficiency. It is important to highlight that due to the low transmission levels of CD and the emergence of other diseases, such as dengue fever, political-administrative questions concerning systematic control programs have been neglected. To maintain these programs, direct support by the municipalities in sustaining their individual programs is necessary, together with the cooperation of the authorities. Unfortunately the National Endemic Control Program of the FUNASA has been discontinued⁸.

The epidemiological surveillance of CD in the central-western region of the State of Minas Gerais, based on knowledge and practice among municipal health agents working in the CD programs of surveillance and control was examined by Villela et al¹⁹. Thirty-five municipalities were visited, involving meetings and the application of a questionnaire pertaining to the knowledge, practice and difficulties of surveillance programs. Analysis of this data showed that 26 agents were employed on temporary (annual) contracts and only two had career stability. Of the agents surveyed, 77.1% commented that the local population had difficulty in identifying the vector insects (triatomines). Twenty-three agents stated that the municipal governments provided adequate conditions for conducting surveillance programs, however 27 mentioned problems, particularly requesting more educational materials, better wages and greater appreciation of their work by the municipal authorities.

In the present study, the species most frequently detected in the dwellings was *Triatoma sordida*, also considered the prevailing species in the State of Mato Grosso do Sul²⁰. *Triatoma sordida* was also the species most frequently captured in the City of Uberlândia. This study is in agreement with research conducted by Silveira et al⁴, who captured and identified in residences, the following species: *Triatoma infestans*, *Triatoma sordida*, *Panstrongylus megistus* and *Rhodnius neglectus*, in the same city. In 2004, Macêdo and Marçal Júnior²¹ registered the presence of *Triatoma sordida* and *Rhodnius neglectus* in dwellings investigated by the authors in the region of

Olhos d'Água, a rural district of Uberlândia. *Triatoma sordida* usually occupies places where *Triatoma infestans* has been eliminated due to the residual effects of the insecticides adopted by the control programs. This species, as well as *Panstrongylus megistus*, has been captured in both wild and urban ecotopes, frequently constituting domestic colonies^{15,22,23}. In other studies, however, this species presented low rates of positivity for *Trypanosoma cruzi*, probably because it prefers to feed on the blood of birds^{1,24}. In the present study, 1.4% of the total of positive triatomine were *Triatoma sordida*, thus constituting the third most prominent species for rates of positivity for *Trypanosoma cruzi*. For this reason *Triatoma sordida* should be the object of continuous monitoring.

In the present study, the primary habitat of *Rhodnius neglectus*, the second most captured species in the intradomicile area, was palm tree leaves¹. The invasion of this wild species into human dwellings, with the risk of transmission of *Trypanosoma cruzi*, may have occurred due to the lack of ecological equilibrium caused by deforestation²⁵.

In Minas Gerais, *Panstrongylus megistus*, second most captured species in the present study, has also been identified as the species most frequently detected in dwellings in studies conducted by Villela et al⁸. *Panstrongylus megistus* is considered to be the vector of the strongest epidemiological importance in Brazil due to its wide geographical distribution, high colonization capacity, domiciliation and adaptation to various sources of food and high susceptibility to *Trypanosoma cruzi*. Following the elimination of *Triatoma infestans* in MG, *Panstrongylus megistus* became the most important vector for CD in the western part of the state, where adult insects, in particular fecundated females, have been captured in the immediate vicinity of houses^{1,8,26}.

In conclusion, there is a need to adapt to new circumstances in epidemiology, with greater emphasis on entomology surveillance, since the potential for adaptation of secondary species of triatomines exists, especially where Chagas' disease is already under control.

This fact corresponds to the renewed risk of the establishment of a new transmission focus, especially in the domicile environment, since these species may occupy the place of the eliminated species. Given these factors, we believe it is important to maintain entomological surveillance in the study area.

CONFLICT OF INTEREST

None of the authors of this paper has a financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of the paper.

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