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Correlation between infection rate of triatominies and Chagas Disease in Southwest of Bahia, Brazil: a warning sign?

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ABSTRACT

Chagas disease, caused by the *Trypanosoma cruzi*, has a wide distribution in South America, and its main method of control is the elimination of triatomines. It is presented here the geographic distribution and the rate of natural infection by *T. cruzi* of triatomines collected and evaluated from 2008 to 2013 in southwest of Bahia. Triatomines were captured in the intradomiciliary and peridomiciliary areas of five cities located in the southwest of Bahia state, identified, and analyzed for the presence of trypanosomatids in their feces. During the study period the number of patients suspected for acute Chagas disease was recovered from the Notifiable Diseases Information System (SINAN). 8966 triatomines were captured and identified as belonging to eight species. Twenty-six presented themselves infected, being *Triatoma sordida* the most abundant and with the highest percentage of infection by *T. cruzi*. Tremedal was the city with the highest number of cases of acute Chagas' disease reported to SINAN. All cities showed triatomines infected with *T. cruzi*, so there is considerable risk of vectorial transmission of Chagas disease in the southwestern Bahia state, evidencing the need for vector transmission control programs and preventive surveillance measures.

Key words: Infection, Chagas disease, *Trypanosoma cruzi*, vector.

INTRODUCTION

The American trypanosomiasis is caused by the flagellate protozoan *Trypanosoma cruzi*. This pathology possesses a wide distribution, being estimated that there are about 7 million people infected by this protozoan and more than 70 million people live in areas of risk of infection. The control of vectors populations is one of the most used methods to prevent the transmission of *T. cruzi* since no vaccine is available to prevent infection and there is no cure during chronic phase (Marin-Neto and Rassi 2009, Mota-Sánchez 2009, WHO 2015).

The Triatominae subfamily was characterized as presenting 143 species that are subdivided into 18 genera and 5 tribes (Galvão et al. 2003, Frías-

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Lasserre 2010). They are characterized by their mouthparts as "pick-suction" that allows them to acquire parasites. Several studies have been conducted to understand the geographical distribution of its species and also their rate of infection (Guzmán-Marín 1990). It has been described different species of Triatominae in Brazil. In Rio Grande do Sul state (located at the south of the country) the most common triatomines are the *Triatoma circummaculata* and the *Triatoma rubrovaria* (Ribeiro et al. 2014); in Minas Gerais (located at the southeastern region), the most common are the *Panstrongylus megistus* and the *Triatoma sordida* (Belisário et al. 2013, Villela et al. 2005); The Brazilian northeast is a region that needs special attention regarding Chagas disease and the presence of vectors in its territory. It is already known that northeastern of Brazil is the second region with the largest triatomine infestation in the country, and the Bahia state in particular has the greatest diversity of species of triatomines in the country (Dias et al. 2000). In Pernambuco state, the species with the highest incidence are *Triatoma pseudomaculata, Triatoma brasiliensis* and the *Pastrongylus lutzi* (Silva et al. 2012). In Ceará state the species found are *Panstrongylus lutzi*, *Triatoma pseudomaculata, T. brasiliensis* and *Rhodnius nasutus* (Caranha et al. 2006, Freitas et al. 2004, 2005, Dias et al. 2008, Sarquis et al. 2004). In Bahia state, the most frequent species found are the *Triatoma sordida, Triatoma sherlocki, Triatoma lenti, Triatoma tibiamaculata, Pastrongylus geniculatus* and also *Triatoma pseudomaculata* (Dias et al. 2000, Santana et al. 2011, Ribeiro et al. 2014). In some regions in Bahia it is also found the *Triatoma infestans,* one of the main vectors of Chagas disease (Araújo et al. 2014, Rassi Jr et al. 2010, 2012).

Different studies have shown several infection rates of triatomine by *T. cruzi.* These infection rates vary from 6.4% (Ribeiro et al. 2014) up to 64.7% of microorganisms similar to *T. cruzi* (Sessa et al. 2002). The natural index of *T. cruzi* infection ranges from 10.8 to 30.2%, depending on species and locations (Sarquis et al. 2004). In Bahia state, a recent study in Salvador and metropolitan area showed a *T. cruzi* infection rate of 54% of all insects analyzed, indicating a high potential risk for parasite transmission (Santana et al. 2011).

Since 2006, Brazil has been found free of vectorial transmission of Chagas disease by *Triatoma infestans*. However, the existence of dwellings whose physical conditions may favor their presence, keeps the risk of this species becoming domiciliated, and, thereby increasing the risk of infection by this route. In Bahia state, the routine actions of control programs are implemented depending on the infestation by the vector species and the epidemiological situation of the disease in the municipalities, defined by the classification according to the degree of risk of transmission. Of the total 417 existing municipalities, 101 (24.2%) municipalities are classified as low risk, 219 (52.5%) of medium risk and 97 (23.3%) high risk.

Although eradication of vectors is somewhat impractical, control programs thereof have shown increasingly effectiveness, and this is represented by the decrease in seroprevalence numbers of Chagas disease in Brazil over the years. In a study conducted from 1975 (year that started the control program of the vectorial transmission of Chagas disease in the country) to 1981, were observed seroprevalence of 8.8% in South and Southeast states, such as Rio Grande do Sul and Minas Gerais, and 5.4% in Bahia (Camargo et al. 1984). In another study, conducted by Ostermayer et al. (2011) in Brazil, was observed a significant decrease in the seroprevalence percentages from 2001 to 2008 in children aged from 0 to 5 years old. In this study, the authors pointed out that the infection would be current, in which positive seroprevalence was 0.03%.

Epidemiological analysis of the distribution of potential vectors for *T. cruzi* is an essential information, in order to have a better understanding of how the transmission of the parasite occurs, and from then on, articulate strategies to control the transmission of Chagas disease (Silva et al. 2012). Therefore, the present work aimed to analyze the distribution scenario of triatomines and their rate of natural infection with *Trypanosoma cruzi* in the southwest region of the state of Bahia from 2008 to 2013.

MATERIALS AND METHODS

COLLECTS

Triatominae were captured, a quarterly frequency, during the period from 2008 to 2013 in the cities of Anagé, Caraíbas, Condeúba, Presidente Jânio Quadros and Tremedal, located in the southwestern region of Bahia (Figure 1). Specimens were collected after active searching manually with forceps and light, inspecting cracks, crevices and hiding places for these triatomines. The collection was carried by endemic control agents of the municipalities under the guidance of technicians from the 20th Regional Directorship of Health, the State Department of Health of Bahia.

TAXONOMIC IDENTIFICATION AND PARASITOLOGICAL RESEARCH

The triatomines were identified and forwarded to Triatomines Laboratory from the core control of endemic diseases of the DIRES20-SESAB (Vitória da Conquista/BA). Over there, the taxonomic identification was held. A parasitological survey was conducted by compressing the abdomen of triatomines and collecting fresh feces, which were used as a sample. These were properly treated and stained with Giemsa for subsequent analysis with an optical microscope, in which was conducted the search by *Trypanosoma cruzi* (Silveira and Sanches 2003). When found even a single individual of *T. cruzi* in triatomine, this was considered infected.

NOTIFICATION OF CHAGAS DISEASE

During the study period, 2008 to 2013, the patients suspected of acute Chagas disease were reported by the Departments of Health in each municipality through the Notifiable Diseases Information System (SINAN).

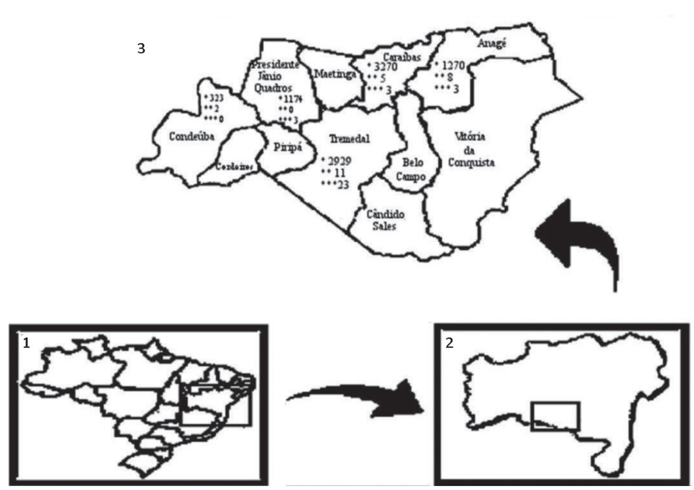

Figure 1 - Geographical distribution map of triatomines in the southwest region of Bahia. **1** – Brazil; **2** – Bahia; **3** – Southwest of Bahia. * - Total number of triatomines; ** - Number of infected triatomines by flagellate protozoa similar to *T. cruzi*. *** - Number of patients with Chagas disease reported by SINAN system, period 2008 to 2013.

RESULTS

A total of 8,966 triatomine from eight different species were collected, being 472 in the intradomiciliary and 8,494 in the peridomiciliary area, among the five cities analyzed within a period of six years (Table I). The species with the highest abundance was the *Triatoma sordida* (8,657/96.55%), followed by *Triatoma pseudomaculata* (129/1.44%), *Pastrongylus geniculatus* (78/0.87%), *Pastrongylus lutzi* (52/0.58%), *Triatoma infestans* (35/0.39%), *Triatoma melanocephala* (12/0.13%), *Rodnius neglectus* (2/0.02%) and *Triatoma vittceps* (1/0.01%) (Table II). Caraíbas was the city with the highest occurrence of triatomines, being captured until the end of this study 3,270 vectors. Soon after, appears Tremedal with 2,929, Anagé with 1,270, Presidente Jânio Quadros with 1,174 and finally Condeúba with the occurrence of 323 triatomine vectors captured (Table I).

As for natural infection of triatomine bugs vectors the results follow. From the total of captured triatomines only 26 (0.29%) showed infection by *T. cruzi* through the parasitological test. Of these vectors, 19 were *T. sordida* (first in frequency in our study) (73.08%) and 7 were *P. geniculatus* (third in frequency) (26.92%). The remaining species showed no *T. cruzi* infection detectable. Among the cities, Tremedal had the highest number of infected triatomines, with 11 vectors (42.31%), these being *P. geniculatus* (7) and *T. sordida* (4). In other cities, only *T. sordida* presented infected vectors, being eight in Anagé (30.77%), five in Caraíbas (19.23%) and two in Condeúba (7.69%). Presidente Jânio Quadros showed no vectors infected by *T. cruzi* according to the parasitological test performed (Table III).

Among all cities, only Condeúba (fifth in occurrence vector) showed no reports of individuals diagnosed with acute Chagas disease in SINAN (Figure 1). In the cities of Anagé, Caraíbas and Presidente Jânio Quadros three cases were reported by each city. Tremedal was the city that had the highest number of acute Chagas disease reports in SINAN, totaling 23 cases. Following the description of years in which cases have been reported (data not shown). In Anagé all cases were reported in 2009. In Caraíbas all in 2013 and in Presidente Jânio Quadros only one case was reported in 2011 and two cases in 2013. In Tremedal, while 2010 was the year in which most reports occurred, with 12 reported cases, one case has been registered in 2008, two cases in 2009, two cases in 2011 and six cases in 2012, there is no recorded cases in the last year of the study (2013).

DISCUSSION

Currently, different strategies to control Chagas disease have been applied. Among them, there is the improvement of housing and surrounding areas in hazardous locations and also methods of sanitary education. All aimed to smaller infestation of triatomines, which are the main responsible vectors for disease transmission (Coura and Dias 2009). In Bahia, of the existing 336 municipalities, only 29 (in the extreme south) were considered without risk of vector transmission. In all others (282) were detected the presence of at least one major vector species (*T. infestans, P. megistus, T. brasiliensis, T. pseudomaculata* and *T. sordida*) (SESAB/DIVEP 2006). In this study, Bahia, obtained from WHO (1991) certification of non-free zone for vector transmission by *T. infestans*, as well as 12 other states, but “possibly interrupted transmission”. Methods based on the control and elimination of the vectors have been already used some decades ago, and were effective in reducing the transmission of Chagas disease, such as the control of *Triatoma infestans* in the late 60s and early 70s (Carvalho et al. 2011).

The present study showed a distribution of vectors in southwestern Bahia similar to the distribution in other regions of Brazil, mainly in the northeast and midwest. The *Triatoma sordida* Stal,

TABLE I
Capture of triatomines per domicile (intra or peri), in municipalities of Bahia – Brazil, 2008 to 2013.

CITY	2008			2009			2010			2011			2012			2013			
	INTRA	PERI	**TOTAL**	INTRA	PERI	**TOTAL**	INTRA	PERI	**TOTAL**	INTRA	PERI	**TOTAL**	INTRA	PERI	**TOTAL**	INTRA	PERI	**TOTAL**	**TOTAL**
ANAGÉ	54	919	**973**	4	70	**74**	0	0	**0**	0	2	**2**	2	82	**84**	3	134	**137**	**1270**
CARAÍBAS	103	2584	**2687**	11	222	**233**	0	169	**169**	0	0	**0**	0	44	**44**	9	128	**137**	**3270**
CONDEÚBA	25	159	**184**	3	26	**29**	0	0	**0**	0	44	**44**	0	24	**24**	2	40	**42**	**323**
P. JÂNIO QUADROS	16	865	**881**	3	19	**22**	0	37	**37**	0	43	**43**	0	16	**16**	16	159	**175**	**1174**
TREMEDAL	**113**	**2062**	**2175**	**0**	**51**	**51**	**32**	**182**	**214**	**1**	**217**	**218**	**0**	**170**	**170**	**75**	**26**	**101**	**2929**
TOTAL	**311**	**6589**	**6900**	**21**	**388**	**409**	**32**	**388**	**420**	**1**	**306**	**307**	**2**	**336**	**338**	**105**	**487**	**592**	**8966**

TABLE II
Occurrence of triatomine vectors captured in municipalities of Bahia – Brazil, 2008-2013.

Vectors*	Anagé						Caraíbas						Condeúba						Pres. Jânio Quadros						Tremedal						Total
	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	
T. sordida	955	72	-	02	80	133	2677	227	169	-	40	136	175	23	-	41	12	28	873	05	27	37	04	151	2158	37	171	178	156	90	8657 (96.55%)
T. psedomaculata	17	01	-	-	01	03	09	06	-	-	04	01	02	02	-	-	06	-	03	16	01	-	03	05	06	02	22	11	08	-	129 (1.44%)
P. geniculatus	-	-	-	-	-	-	01	-	-	-	-	-	06	02	-	02	05	12	01	-	-	03	-	08	08	10	07	06	04	03	78 (0.87%)
P. lutzi	01	01	-	-	03	01	-	-	-	-	-	-	01	01	-	01	01	01	01	-	09	03	03	07	01	02	-	05	02	08	52 (0.58%)
T. infestans	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	01	-	02	-	14	18	-	-	35 (0.39%)
T. melanocephala	-	-	-	-	-	-	-	-	-	-	-	-	-	01	-	-	-	-	03	01	-	-	05	02	-	-	-	-	-	-	12 (0.13%)
R. neglectus	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	02	-	-	-	-	-	-	02 (0.02%)

TABLE II (continuation)

Vectors*	Anagé						Caraíbas						Condeúba						Pres. Jânio Quadros						Tremedal						**Total**
	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	
T. vitticeps	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	**01**	-	-	-	-	-	-	-	-	-	-	-	-	**01 (0.01%)**
Total	1270						3270						323						1174						2929						**8966**

* The identification of vectors occurred in Laboratory of triatomines from the Core control of endemic diseases of the 20th Regional Directorate of Health – Vitória da Conquista/ BA-Brazil.

TABLE III

Occurrence of triatomine vectors infected by *Trypanosoma cruzi* in municipalities of Bahia – Brazil, 2008-2013.

Vectors*	*T. sordida*						*P. geniculatus*						**Total**
	2008	2009	2010	2011	2012	2013	2008	2009	2010	2011	2012	2013	
Anagé	**01**	**02**	-	**01**	**02**	**02**	-	-	-	-	-	-	**8 (30.77%)**
Caraíbas	**04**	-	**01**	-	-	-	-	-	-	-	-	-	**5 (19.23%)**
Condeúba	-	**02**	-	-	-	-	-	-	-	-	-	-	**2 (7.69%)**
Pres. Jânio Quadros	-	-	-	-	-	-	-	-	-	-	-	-	-
Tremedal	-	-	**01**	-	**03**	-	-	-	**07**	-	-	-	**11 (42.31%)**
Total	**19 (73.08%)**						**07 (26.92%)**						**26**

* The identification of vectors occurred in Laboratory of triatomines from the Core control of endemic diseases of the 20th Regional Directorate of Health – Vitória da Conquista/ BA-Brazil.
The vectors *T. infestans*, *T. pseudomaculata, P. lutzi, T. melanocephala, T. vitticeps,* and *R. neglectus* captured were also investigated for infection by *T. cruzi.* The results for these vectors were all negative.

1859 (species with the highest occurrence in this study) showed also to be quite present in several states. According to Gurgel-Gonçalves et al. (2012), who conducted a study based on ecological niche model, *T. sordida* showed greater distribution in the Brazilian Cerrado, and it is present in virtually all the northeast, beyond the southeast and Brazilian central-western. Moreover, this appears to be the vector with the highest occurrence not only in this study (which analyzed southwestern Bahia) but also in other studies such as in the state of Goiás, where between the years 2000 and 2003 among 51,570 triatomines captured more than 86% (44,374) were from this species (Oliveira and Silva 2007). Another relevant factor regarding the *T. sordida* is that some of these insects already have been found resistant to deltamethrin, one of the chemicals used in programs to combat vectors (Pessoa et al. 2014).

The risk of transmission of the parasite by the *T. sordida* is considered low, since this usually does not dwell intradomicile regions and can be found more often in the peridomicile (Forattini et al. 1979). However, this is also more related to agricultural environments which require attention and control of this vector, since the cities analyzed consist of cities with agricultural activities; no major industrial centers; and there are still people who survive from the field work (Klink and Machado 2005). Similar to these studies, the frequency of *T. sordida* found by our work involves mainly agricultural counties.

The *Triatoma pseudomaculata* Corrêa & Espinola, 1964 is considered one of the major triatomine in the northeast of the country, since it is present in all states of the region and has been considered the most prevalent vector in several northeastern states. For several years, this region was the epicenter of its origin and dispersion, and studies suggest that from the northeast these vectors infested other states, such as Minas Gerais and Goiás (Dias et al. 2000, Silveira et al. 2002). This species are generally present in peridomicile areas, dwelling mainly natural, being present generally in lofts, fences, roof spaces, tree holes and piles of firewood or bricks (Assis et al. 2007). However, in the specimens captured in intradomiciliary environments during our study, no infection has been observed by *T. cruzi*. Studies show that their presence has low conversion rates for metacyclic trypomastigotes, and in this way, *T. pseudomaculata* have not been considered an important vector of Chagas disease (Forattini et al. 1981, Perlowagora-Szumlewicz and Moreira 1994).

Other species in our study that presented infection by *T. cruzi* was *Pastrongylus geniculatus* Latreille, 1811, which presents a wide distribution throughout the country, but not restricted only to this, being widely distributed by Americas and present at least in 16 countries (Leite et al. 2007). Despite the highest rate of infection among the species analyzed in this study originally inhabits in the wild environment, and has not been considered such an important vector of *T. cruzi* because their bite is painful and cause allergic reactions, which hinders their efficiency of vector transmission (Diotaiuti 2009), it becomes necessary to take special care of this vector in these regions, since according to Marçal and Macêdo (2004), this species is migrating to the intradomiciliary regions.

Triatoma infestans Klug, 1834 was found in considerable numbers in our study, mainly in the city of Tremedal. In the early 1990s, this vector has demonstrated a geographic distribution of approximately 37,282.27 mi and was responsible for the transmission of Chagas disease to about 9 million people (WHO 1991). It is noteworthy that *Triatoma infestans* was once considered one of the main vectors of Chagas disease in Brazil, mainly in the state of São Paulo. However, more recently, some states have already received certificate of disposal of this vector (Carvalho et al. 2011, Oliveira and Silva 2007). Older studies, such as Barrett et al. (1979) showed a large amount of this vector in the southwestern region of Bahia, both intradomicile and peridomicile, with 5% infection rate, providing a high risk of transmission due to its efficiency as a vector. However, in 2006, Brazil received an International certification of elimination of transmission of Chagas disease by *T. infestans*, granted by the Pan American Health Organization (PAH), joining Uruguay and Chile, certified in 1997 and 1999, respectively (Schofield et al. 2006, Silveira and Dias 2011).

Despite the few cases of diagnosed patients reported, this study shows the possibility of a relationship between the number of infected triatomine bugs and the number of people reported with Chagas disease, since Tremedal was the city with the greatest number of insects infected and also with the highest number of cases reported in SINAN of individuals diagnosed with acute Chagas disease. However, in a study by Carvalho et al. (2011), it was observed that despite the high rate of infected triatomine, 50% positive for *T. cruzi*, in human only 1.2% were positive showing no direct relationship between the presence of infected triatomine vector and transmission to humans. Although, this same study observed that the average distance of triatomines collection points from the housing of individuals analyzed were 257.76 yd and have not being considered peridomicile region, also, becoming a difficulty to the contact between humans and vectors in that region. Although our study makes no correlation between the housing of patients with acute disease and the distance from the site of capture of the insects, the cities where the data collections were performed are primarily agricultural.

Therefore, due to the capture of triatomines infected with *T. cruzi* in almost all the cities analyzed in this study, it was observed that in the southwestern region of Bahia there is a considerable risk of vector transmission of this parasite. Although they are considered less efficient species vectors than *T. infestans,* we reported *T. sordida* and *P. geniculatus* also infected. These data indicates that more attention should be given and new strategies should be developed to control *T. cruzi* vectors in this region. This study was the first to show a clear need of entomological surveillance and monitoring of these potentially hazardous locations, beyond the awareness of the population about the possible locations of vectors infestation. However, more studies are necessary over the next few years to analyze if the sanitary measures taken are effective and evaluate new methods of control.

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RESUMO

A doença de Chagas, causada pelo *Trypanosoma cruzi*, possui uma ampla distribuição na América do Sul, e tem como principal método de controle a eliminação de triatomíneos. Apresenta-se aqui a distribuição geográfica e a taxa de infecção natural por *T. cruzi* de triatomíneos coletados e avaliados de 2008 a 2013, no sudoeste da Bahia. Os triatomíneos foram capturados nas áreas intradomiciliares e peridomiciliares de cinco cidades localizadas no sudoeste da Bahia, identificados e analisados quanto à presença de tripanossomatídeos em suas fezes. Durante o período de estudo o número de pacientes com suspeita de doença de Chagas aguda foi recuperado a partir do Sistema de Informação de Agravos de Notificação (SINAN). 8966 triatomíneos foram capturados e identificados como pertencentes a oito espécies. Vinte e seis apresentaram-se infectados, sendo *Triatoma sordida* o que mostrou maior percentagem de infecção pelo *T. cruzi*. Tremedal foi a cidade com o maior número de casos de doença de Chagas aguda relatado pelo SINAN. Todas as cidades apresentaram triatomíneos infectados com o *T. cruzi*,

por isso, existe um risco considerável de transmissão vetorial da doença de Chagas no sudoeste da Bahia, evidenciando a necessidade de programas de controle de vetores de transmissão e medidas de vigilância preventiva.

Palavras-chave: infecção, doença de Chagas, *Trypanosoma cruzi*, vetor.

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