Reassessing the entomological investigation around the first autochthonous case of Chagas disease in Western Brazilian Amazon

Nelson Ferreira Fé[1], Menabarreto Segadilha França[2], Filipe Anibal Carvalho-Costa[3]/+

[1]Gerência de Entomologia, Fundação de Medicina Tropical do Amazonas, Manaus, AM, Brasil [2]Faculdade de Medicina, Universidade Federal do Amazonas, Manaus, AM, Brasil [3]Laboratório de Sistemática Bioquímica, Instituto Oswaldo Cruz-Fiocruz, Av. Brasil 4365, 21045-900 Rio de Janeiro, RJ, Brasil

In 1979, the first autochthonous case of Chagas disease in the Western Brazilian Amazon was reported and an entomological survey was carried out around it. Specimens of Rhodnius pictipes *and* Rhodnius robustus *were collected in intradomicile and sylvatic ecotopes. Adult bugs were infected with trypanosomatids. Invasion of houses by triatomines was demonstrated and the presence of infected bugs inside dwellings was associated with the possibility of vector-borne Chagas disease. Continuous entomological surveillance employing additional taxonomic tools is needed in the Brazilian Amazon in order to better understand the dynamics of house invasion by sylvatic triatomines and the risk of* Trypanosoma cruzi *infection transmission.*

Key words: Chagas disease - triatomines - Amazon

Traditionally considered hypoendemic in the Brazilian Amazon, Chagas disease is now regarded as an emergent and neglected condition in this region (PAHO 2005).

In 1979, the first autochthonous case of Chagas disease in the Western Brazilian Amazon - which includes the states of Acre, Roraima, Rondônia and Amazonas (AM) - was accidentally identified in the municipality of São Paulo de Olivença, AM, in a blood smear performed in order to diagnose malaria (França et al. 1980). The patient was an acutely ill four-year-old girl. Prior to this report, eight parasitologically confirmed cases were described in the Eastern Amazon, mainly in state of Pará (PA) (Shaw et al. 1969, Silveira et al. 1979). Over the last few decades, nearly 300 acute cases associated with outbreaks (probably associated with *Trypanosoma cruzi*-contaminated beverages) and over 125 acute cases without any relationship to those outbreaks have been reported in the Amazon (Aguilar et al. 2007).

Various triatomines species have been collected in a wide variety of natural ecotopes throughout Amazonian Brazil, with high rates of infection by *T. cruzi* (Abad-Franch & Monteiro 2007, Aguilar et al. 2007). Here, we describe and reassess the entomological investigation around the first case of Chagas disease described in the Western Brazilian Amazon.

São Paulo de Olivença is situated in the Alto Solimões Region (3.37ºS 68.87ºW), 1,432 km away from Manaus, the state capital. About 30,000 people inhabit the municipality. A portion of the population is dispersed in small riverine communities like Boa Esperança, where the Chagas disease case was reported in October 1979 and this survey was carried out six months later, in April/May 1980. People living in Boa Esperança practice subsistence agriculture, fishing and gathering of forest products like rubber and nuts. Houses are rudimentary (made of straw and wood), with incomplete walls that permit the entry of insects. Dwellings are situated very near the forest, from which the inhabitants gather food and domestic utensils.

All dwellings (n = 12) in this community were investigated. Collections were performed using the traditional search and capture method with the prior consent of inhabitants in three areas: (i) inside houses (intradomicile), sometimes following application of the dislodging agent Piriza® 0540 - 1%; (ii) around houses (chicken huts, piles of wood, rubber smokers, animal holes e.g., defined as extradomicile); and (iii) in palm trees surrounding houses (sylvatic/peridomicile). One specimen from each species of palm tree was sacrificed and dissected with the prior consent of landowners. Approximate distances from the trees to the surveyed houses were as follows: *Astrocarium murumuru* (murumuru), 100 m; *Attalea phalerata* (uricuri, acuri), 10 m; *Mauritia carana* (caraná), 100 m; *Mauritia flexuosa* (buriti), 50 m.

As presented in the Table, triatomines of different developmental stages were found. Twenty-two specimens of *Rhodnius pictipes* were collected (12 in intradomicile and 10 in sylvatic/peridomicile ecotopes) and 25 specimens of *Rhodnius robustus* were captured (2 in intradomicile and 23 in sylvatic/peridomicile ecotopes) ($p < 0.001$; chi-square test). Interestingly, five adult triatomines were collected inside bed nets used for protection against mosquitoes (3 *R. pictipes* and 2 *R. robustus*). The presence of flagellated trypanosomatids in adult bugs was assessed by simple examination of faeces obtained by dissection of the posterior intestine of the insect. All adult triatomines were infected.

Although 86% of triatomines collected in intradomicile areas were *R. pictipes*, both species were found inside dwellings. People living in Boa Esperança seem to be familiar with triatomines, as nicknames such as "Jurupari" (evil spirit) and "Camareta" (characteristic smell) were attributed to them.

+ Corresponding author: guaratiba@ioc.fiocruz.br
Received 22 September 2008
Accepted 6 January 2009

In the Amazon, house invasion by sylvatic triatomines is considered a risk for vector-borne Chagas disease (Coura et al. 1994, 1999). Sylvatic triatomines of the genus *Rhodnius* fly into houses, maintaining the risk of *T. cruzi* transmission (Coura et al. 1999). Determination of the specific status of *Rhodnius* specimens collected inside houses in the Amazon is sometimes difficult and epidemiologically important and distinct Chagas disease transmission potentials remain to be elucidated between species. Many tools have been utilized in order to clarify this issue, including traditional and geometric morphometrics and molecular genetic techniques (Villegas et al. 2002, Pavan & Monteiro 2007, Harry et al. 2008).

Monteiro et al. (2003) emphasized the morphological and geographical overlap between *R. robustus* and *Rhodnius prolixus,* identifying the possibility of misidentifications. Based on molecular phylogenetics, these authors demonstrated the relationships between these species, showing that *R. robustus* from the Orinoco Basin is more closely related to *R. prolixus* than to the other *R. robustus* specimens from the Amazon region.

Recently, the specific status of sylvatic triatomines collected inside houses in Amazonian Venezuela was assessed suggesting the presence of *R. prolixus* (but not *R. robustus*) in palm trees and demonstrating its potential to invade and colonize houses (Fitzpatrick et al. 2008).

Although our results are temporally and geographically limited, we show that the main species inhabiting palm trees in Boa Esperança, based on morphological characteristics, is probably *R. robustus*. Although this species is considered of minor epidemiological importance for *T. cruzi* transmission, two adult *R. robustus* females were found invading houses; they were inside bed nets and both were infected with flagellate trypanosomatids. Feliciangeli et al. (2002) demonstrated that *R. robustus*, which is well differentiated from *R. prolixus* based on random amplified polymorphic DNA, invades houses in Venezuela and feeds on humans or bites people outdoors. In this context, the authors argue that the lack of triatomines inside houses could mean that they leave houses after feeding or die without reproducing therein.

According to the geographical distribution of Amazonian triatomines reviewed by Abad-Franch and Monteiro (2007) and the genotypic classification proposed by Monteiro et al. (2003), *R. robustus* belonging to genotypes II, III and IV (but not *R. prolixus*) are present in the biogeographical province of Madeira, where our study was carried out.

In the present study, the main species found inside houses was *R. pictipes*. This species has a broad geographical distribution throughout the Amazon (Carcavallo et al. 1999). Lainson et al. (1979) and Miles et al. (1983) demonstrated the presence of *R. pictipes* infected with *T. cruzi* in three different species of palm trees, demonstrating that they are frequently attracted into houses by light. This represents a potential source of Chagas disease. Furthermore, the epidemiological importance of *R. pictipes* was demonstrated in Equador by Aguilar et al. (1999).

The risk of Chagas disease transmission in the Amazon is, therefore, enhanced by the presence of palm trees near households. Teixeira et al. (2001) described a complete trophic network comprising different species dwelling in palm trees microhabitats. These include distinct families of insecta (including triatomines), amphibians, birds and marsupials, like *Didelphis marsupialis*, a sylvatic reservoir of *T. cruzi*. The importance of palm trees in the ecology of sylvatic triatomines in PA was demonstrated by Valente et al. (1998). In addition, the close relationship between wild triatomines and humans has

TABLE

Triatomines collected in São Paulo de Olivença, state of Amazonas, 1980

	Sylvatic/peridomicile		Intradomicile	
Triatomine species and stages	*Astrocarium murumuru* (murumuru)	*Athalea phalerata* (acuri)	Inside bed nets	Floor/roof /walls
Rhodnius robustus				
1st insthar nymph	2	3	-	-
2nd insthar nymph	3	-	-	-
3rd insthar nymph	-	5	-	-
4th insthar nymph	4	3	-	-
5th insthar nymph	-	1	-	-
Adult female	-	1	2	-
Adult male	-	1	-	-
Rhodnius pictipes				
1st insthar nymph	-	-	-	-
2nd insthar nymph	-	2	-	-
3rd insthar nymph	-	4	-	-
4th insthar nymph	-	-	-	-
5th insthar nymph	-	-	-	-
Adult female	-	2	3	6
Adult male	-	2	-	3

been associated with contamination of food with *T. cruzi*, leading to many outbreaks of orally transmitted Chagas disease (PAHO 2006). It is important to emphasize that different species of palm trees represent a very important economic resource in the Amazon. Riverine communities collect fruits and seeds in order to prepare beverages and oils and leaves to produce several domestic facilities.

Controlling *T. cruzi* transmission in such an eco-epidemiological scenario may be a challenge. Chagas disease transmission has been treated as a major public health problem in the Brazilian Amazon and relates to a complex framework of cultural and biological determinants. Implementation of entomological and active epidemiological surveillance, together with the identification and treatment of acute cases (which can take advantage of the extensive laboratorial network involved in malaria control throughout the Amazon) is urgently needed.

REFERENCES

Abad-Franch F, Monteiro FA 2007. Biogeography and evolution of Amazonian triatomines (Heteroptera: Reduviidae): implications for Chagas disease surveillance in humid forest ecoregions. *Mem Inst Oswaldo Cruz 102* (Suppl. I): 57-70.

Aguilar HM, Abad-Franch F, Dias JC, Junqueira AC, Coura JR 2007. Chagas disease in the Amazon region. *Mem Inst Oswaldo Cruz 102* (Suppl. I): 47-56.

Aguilar VHM, Abad-Franch F, Racines VJ, Paucar CA 1999. Epidemiology of Chagas disease in Ecuador. A brief review. *Mem Inst Oswaldo Cruz 94* (Suppl. I): 387-393.

Carcavallo RU, Curto de Casas SI, Sherlock IA, Girón IG, Jurberg J, Galvão C, Mena Segura CA, Noireau F 1999. Geographical distribution and alti-latitudinal dispersion. In RU Carcavallo, I Galindez Girón, J Jurberg, H Lent (eds.), *Atlas of Chagas Disease Vectors in the Americas*, Vol. 3, Editora Fiocruz, Rio de Janeiro, p. 747-792.

Coura JR, Barret TV, Naranjo CM 1994. Ataque de populações humanas por triatomíneos silvestres no Amazonas: uma nova forma de transmissão da infecção Chagásica? *Rev Soc Bras Med Trop 27*: 251-253.

Coura JR, Junqueira AC, Boia MN, Fernandes O 1999. Chagas disease: from bush to huts and houses. Is it the case of the Brazilian Amazon? *Mem Inst Oswaldo Cruz 94* (Suppl. I): 379-384.

Feliciangeli MD, Dujardin JP, Bastrenta B, Mazzarri M, Villegas J, Flores M, Muñoz M 2002. Is *Rhodnius robustus* (Hemiptera: Reduviidae) responsible for Chagas disease transmission in Western Venezuela? *Trop Med Int Health 7*: 280-287.

Fitzpatrick S, Feliciangeli MD, Sanchez-Martin MJ, Monteiro FA, Miles MA 2008. Molecular genetics reveal that sylvatic *Rhodnius prolixus* do colonise rural houses. *PLoS Negl Trop Dis 2*: e210.

França MS, Frade JM, Konazugawa K, Almeida FB 1980. Doença de Chagas: primeiro caso autóctone na Amazônia Ocidental, Amazonas, Brasil. *Acta amaz (Manaus) 10*: 759-762.

Harry M, Roose CL, Vautrin D, Noireau F, Romaña CA, Solignac M 2008. Microsatellite markers from the Chagas disease vector, *Rhodnius prolixus* (Hemiptera: Reduviidae), and their applicability to *Rhodnius* species. *Infect Genet Evol 8*: 381-385.

Lainson R, Shaw JJ, Fraiha H, Miles MA, Draper CC 1979. Chagas's disease in the Amazon Basin: 1. *Trypanosoma cruzi* infections in sylvatic mammals, triatomine bugs and man in the State of Pará, North Brazil. *Trans R Soc Trop Med Hyg 73*: 193-204.

Miles MA, Arias JR, de Souza AA 1983. Chagas disease in the Amazon Basin: V. Periurban palms as habitats of *Rhodnius robustus* and *Rhodnius pictipes*-triatomine vectors of Chagas disease. *Mem Inst Oswaldo Cruz 78*: 391-398.

Monteiro FA, Barrett TV, Fitzpatrick S, Cordon-Rosales C, Feliciangeli D, Beard CB 2003. Molecular phylogeography of the Amazonian Chagas disease vectors *Rhodnius prolixus* and *R. robustus*. *Mol Ecol 12*: 997-1006.

PAHO - Pan American Health Organization 2005. Implementation of the Intergovernmental Initiative for Surveillance and Control of Chagas Disease in the Amazon Region (AMCHA). International Meeting on Surveillance and Prevention of Chagas Disease in the Amazon Region (Manaus, Amazonas State, Brazil, 19-22 September 2004). Document OPS/DPC/ CD/321/05, 16 pp. Available from: http://www.paho.org/english/AD/DPC/CD/dch-amcha-2004.pdf.

PAHO - Pan American Health Organization 2006. Informe de la Consulta Técnica en Epidemiología, Prevención y Manejo de la Transmisión de la Enfermedad de Chagas como Enfermedad Transmitida por Alimentos (ETA), Rio de Janeiro, Brasil, Mayo 2006, 46 pp. Available from: http://bvs.panaftosa.org.br/textoc/informe_eta.pdf.

Pavan MG, Monteiro FA 2007. A multiplex PCR assay that separates *Rhodnius prolixus* from members of the *Rhodnius robustus* cryptic species complex (Hemiptera: Reduviidae). *Trop Med Int Health 12*: 751-758.

Shaw JJ, Lainson R, Fraiha H 1969. Considerações sobre a epidemiologia dos primeiros casos autóctones de doença de Chagas registrados em Belém, Pará, Brasil. *Rev Saude Publica 3*: 153-157.

Silveira PT, Dias MGV, Pardal PP, Lobão AO, Melo GB 1979. Novo caso autóctone de doença de Chagas registrado no Estado do Pará, Brasil. *Hiléia méd (Belém) 1*: 61-62.

Teixeira AR, Monteiro PS, Rebelo JM, Argañaraz ER, Vieira D, Lauria-Pires L, Nascimento R, Vexenat CA, Silva AR, Ault SK, Costa JM 2001. Emerging Chagas disease: trophic network and cycle of transmission of *Trypanosoma cruzi* from palm trees in the Amazon. *Emerg Infect Dis 7*: 100-112.

Valente VC, Valente SA, Noireau F, Carrasco HJ, Miles MA 1998. Chagas disease in the Amazon Basin: association of Panstrongylus geniculatus (Hemiptera: Reduviidae) with domestic pigs. *J Med Entomol 35*: 99-103.

Villegas J, Feliciangeli MD, Dujardin JP 2002. Wing shape divergence between *Rhodnius prolixus* from Cojedes (Venezuela) and *Rhodnius robustus* from Mérida (Venezuela). *Infect Genet Evol 2*: 121-128.