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### **Major Article**

# Factors associated with the presence of triatomines in rural areas of south Argentine Chaco

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### **Abstract**

**Introduction:** The domestic and peridomestic presence of *Triatoma infestans* depends on several factors, such as human behavior, vector behavior, ecology, and the environment. **Methods:** This work was conducted in 139 domiciliary units, where triatomines were captured and risk factors in domiciles and peridomicilies were recorded. Household dwellers were interviewed to obtain information about practices regarding this disease and entomological indicators were calculated. **Results:** Infestation indices were 59.7% for house compounds, 4.3% for domestic areas and 58.3% for the peridomestic areas. Intradomicile infestation was significantly associated with housing characteristics. The presence of chicken coops in peridomicilies was associated with an increased risk of infestation. Of the respondents, 80% did not recognize the importance of the peridomiciliary structures for triatomine control and had infested peridomicilies. **Conclusions:** The results show the importance of peridomiciles as refuge sites for Triatominae bugs; however, household dwellers do not perceive peridomiciles as areas that favor the presence of vectors. Actions for raising awareness about factors that favor the presence of triatomines are needed to improve the conditions of peridomiciliary environments.

**Keywords:** Chagas disease. Risk factors. *Triatoma infestans*.

### INTRODUCTION

Chagas disease is the most important infection transmitted by vectors in Latin America, affecting 21 countries, including Argentina<sup>1</sup>. In vast areas of these countries, rural populations live in precarious houses that favor colonization by vectors. This disease is caused by the protozoa *Trypanosoma cruzi*, which is transmitted to humans through the feces of infected blood-sucking insects belonging to the subfamily Triatominae (Heteroptera: Reduviidae).

Triatoma infestans (Klug) is the main vector of Chagas disease in the southern cone of South America. In these countries, control measures implemented by INCOSUR since 1991 have succeeded in reducing the original range of this

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vector by approximately 80%, and in interrupting vectorial transmission by *T. infestans* in Brazil, Chile, and Uruguay<sup>2,3</sup>. However, there are persistent populations of triatomines in houses and peridomiciles, especially in the eco-region of the Gran Chaco, a semi-arid plain of 1.1 million km<sup>2</sup>, which covers south-eastern Bolivia and much of central and northern Argentina, to the extreme northwest of Córdoba province<sup>4,5,6,7</sup>. Paraguay recently received certification as a "free country of home vector transmission of Trypanosoma cruzi" as verified by an international mission of independent experts convened by the Pan American Health Organization<sup>8</sup>. The presence of triatomines in households depends on a number of factors related to the building materials and construction conditions of the dwellings and certain peridomiciliary structures (goat and pig corrals plus chicken coops) that provide abundant refuge sites for these insects. The presence of triatomines is also related to the householders' cultural patterns9,10. Rural areas within the Gran Chaco have been the target of several studies related to the identification of these risk factors, such as houses with adobe

and/or cracked walls, dirt floors, thatched roofs, precarious hygienic conditions, easy access to food supplies, and dogs and chickens sleeping inside or near the house. The presence of these factors increases the risk of triatomine infestation in these rural areas<sup>11,12</sup>.

In addition, several studies indicate that the structural characteristics of the peridomiciliary annexes, such as the materials used for construction<sup>13,14</sup>, as well as housing proximity<sup>15</sup>, pose different risk levels of re-infestation of houses<sup>16,17</sup>, which make control actions more difficult to complete. The persistence of *T. infestans* in this region might be caused by the important role of peridomiciles as the main source of indoor re-infestation<sup>6,18,19</sup>. Human behavior can also influence intradomicile and/or peridomiciliary infestation. Precarious hygienic conditions contribute to the maintenance of triatomines in the intradomicile. Other issues such as the accumulation of firewood can promote passive transport of triatomines inside dwellings<sup>20,21</sup>.

Risk factors related to the domestic presence of triatomines may vary between regions, because of the differences in human behavior, vector behavior, ecology, and environment. Therefore, a more general perspective is needed to simultaneously consider the social, economic, and biological processes at the local and, if necessary regional levels, to achieve effective and sustainable vector and disease control<sup>22</sup>.

The northwest area of Córdoba province, Argentina, in the extreme south of the Gran Chaco region, has been historically endemic to Chagas disease and shows a heterogeneous scenario of *T. cruzi* (Chagas) transmission that can be attributed to differences in vector control interventions, changes in land use, and socioeconomic factors in the last decades<sup>23,24</sup>. The Report of the National Chagas Program<sup>25</sup> included Córdoba among the provinces with a medium risk level of vector transmission of *T. cruzi* due to insufficient vector control and poor coverage of control campaigns. However, there are no entomological data available that allow us to explain the persistence of triatomines in this region nor have peridomiciles of these areas been characterized.

Given the importance of identifying the main risk factors in each region, this work aims to describe, identify, and analyze possible risk factors associated with peridomestic and domestic infestation by triatomines in northwestern Córdoba.

### **METHODS**

### Study area

The study was conducted in rural communities of Cruz del Eje (30° 44′ 04″ S, 64° 47′ 57″ W), Córdoba province, northwestern Argentina, between December 2012 and November 2013 (**Figure 1**). This area is located within the phytogeographical Chaco province, which is characterized by

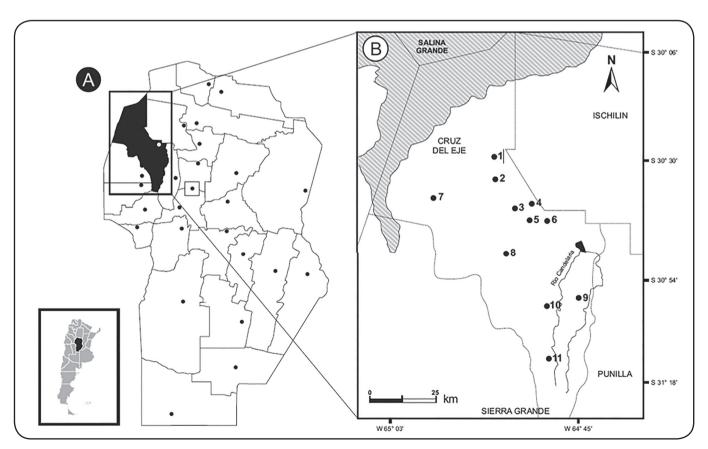


FIGURE 1: A: Location of Cruz del Eje, Córdoba province, Argentina. B: Map of the study area. Rural communities (dots): 1: Villa Luján, 2: Puesto Torrado, 3: Palo Parado, 4: El Simbolar, 5: El Barrial, 6: El Brete, 7: Las Casillas, 8: Villa de Soto, 9: Aguas Coloradas, 10: Cruz de Caña, 11: Ciénaga de Britos.

subtropical dry climate with a summer season from October to March. The average monthly temperature is 26 °C, and 70% of the rain occurs mainly from November to February<sup>26</sup>.

A cross-sectional, observational, and descriptive study was conducted in the domiciliary unit, which included the dwelling (in-home) and the associated peridomicile (environment around the dwelling where the residents conduct their daily routine activities). In the peridomiciliary area, there are usually annexes contain some types of hosts, including, chickens, goats, horses, and pigs. Only animals raised in pens were considered; loose animals in the peridomiciliary area (e.g. hens resting under trees) were not included in the study. The sites and the houses visited were selected according to the recommendations of the National and Provincial Program of Chagas. We visited 139 domiciliary units, which had not been sprayed for more than three years. The location of each domiciliary unit was recorded using a GPS (Garmin eTrex 20). In each domiciliary unit, the type of peridomiciliary annex was recorded considering the host (chicken coop, goat, and pigs corrals, etc.), structure, and distance (m) to the house. Inside the house and each peridomiciliary annex, a search was conducted to locate and capture triatomines, current risk factors were identified, and dwellers were interviewed to obtain demographic data as well as information about their knowledge on Chagas disease and related practices.

All houses were visited by Chagas Disease Program staff. Data collection tools used were previously validated by the staff. The inclusion criteria for the participation of inhabitants in the interview were: being permanent residents of the visited house, being over 18 years old, and voluntarily agreeing to participate in the study after being informed of the purposes.

### **Triatomine surveys**

The hour-man technique<sup>27</sup> was used for the search of triatomines inside the dwelling and in the peridomiciliary annexes. In the laboratory, triatomines were identified according to stage of development, sex and species, and the presence of *T. cruzi* in insect feces was analyzed using a conventional Zeiss optical microscope with the 10x and 40x objectives. Taxonomic identification was performed by observing the external morphology according to Lent & Wigodzinsky<sup>28</sup> and Jurberg et al.<sup>29</sup> for adults and Brewer et al.<sup>30</sup> for nymphs.

The following entomological indexes were calculated: Domiciliary infestation index (DI) =  $100 \times$  (number (n) of houses infested/n houses examined). Peridomiciliary infestation index (PI) =  $100 \times$  (n° of peridomiciles infested/total n of peridomiciles examined). Natural Infection Index (NI): n° of triatomine infected by *T. cruzi/*n° of Triatomine examined x 100. House unit infestation index (HII) =  $100 \times$  (n of infected domiciliary units/n of houses examined).

# Household Survey: risk factors, knowledge, and practice

The house and peridomicile characteristics evaluated as risk factors were selected based on previous works<sup>9,15,30</sup>. In each unit, five risk factors related to building materials, dwelling features,

and peridomiciliary annexes were considered (**Table 1**). Each house was classified according to the type of construction as vulnerable, or not vulnerable, to colonization. The houses considered vulnerable had unplastered concrete roofs, or brick roofs or had thatched roofs with unplastered walls. Likewise, the chicken coops were classified as vulnerable, or not vulnerable, to colonization according to the criteria of Ortiz et al.<sup>14</sup>.

To assess residents' knowledge and practices, we used an interview model designed with the Chagas National Program, based on a list of basic notions<sup>9</sup>.

A total of 10 items associated with knowledge and practices were defined (**Table 2**).

### **Statistical Analysis**

Data were analyzed in two stages: a descriptive one, in which basic statistics were calculated (proportions, average values), and an inferential one, in which the individual effects of categorical independent variables (rates of infestation) were associated with the dependent variables (type of house, features of the coops, knowledge) through contingency tables. The Odds Ratios (OR) and their respective confidence intervals (95% CI) were estimated (SPSS vs 12).

#### **RESULTS**

All rural communities were found to be positive for the presence of triatomines in the domiciliary units. A total number of 1135 triatomines were collected, 74 (6.5%) of which were captured in houses (10 adults and 64 nymphs) and 1061 (93.5%) in peridomiciles (390 adults and nymphs 671). Most (99%) of the insects collected were *T. infestans*, with the remaining ones being four adults of *Triatoma guasayana* (Wygodzinsky & Abalos), and four adults of *Triatoma garciabesi* (Jurberg et al.29), which were captured in peridomiciles. Of the total collected triatomines, only four adults of *T. infestans* were infected with *T. cruzi* (NI: 0.35) The house unit infestation index (HII) was 59.7% (83/139), the domiciliary infestation index (DI) was 4.3% (6/139), and the peridomiciliary infestation index (PI) was 58.3% (81/139).

### Analysis of risk factors associated with the presence of triatomines in households

Of the 139 houses visited, 104 were included in the analysis, because inhabitants of some houses were absent or were reluctant to participate, preventing us from characterizing those houses. Of all houses evaluated, 61.1% had one or more of the risk factors considered in this study; of those houses, 26% (27/104) were classified as vulnerable to colonization (**Table 3**). Household infestation was significantly associated with housing characteristics. The contingency tables showed that 80% of the houses with a wall suitable for triatomine refuge were positive for the presence of triatomines, and 60% of the houses with a roof suitable for triatomine refuge were infested. A house vulnerable to colonization had a 13.21 times higher probability of being infested than a non-vulnerable house (p <= 0.05).

Houses with unplastered walls, even those with concrete roofs, had a 20.7 times greater risk of infestation (p < 0.001)

TABLE 1: Description of risk factors associated with the presence of triatomines in rural communities of Cruz del Eje, Córdoba province, Argentina.

Risk factor	Description
Roofs providing suitable refuge for triatomine	Unplastered brick or concrete, straw or cane thatched roofs <sup>21,33</sup>
2. Walls providing suitable refuge for triatomine	Adobe, unplastered wall <sup>32</sup>
3. Animals sleeping inside the house	Dog/chickens
4. Chicken coops vulnerable to colonization	Bricks, cardboard, fabrics, billets <sup>14,18,21</sup>
5. Chicken coops less than 12 m away from the house <sup>17</sup>	

than those with uncracked plastered walls. Similarly, in houses that had roofs at risk of infestation (unplastered concrete or brick, or thatched roofs) the risk was 7.23 times greater than in houses with plastered roofs (p < 0.001) (**Table 3**).

## Analysis of risk factors associated with the presence of triatomines in the peridomicile

Of all houses visited, 85.6% (119/139) had some type of annex in the peridomiciles, the most common ones being chicken coops (78.4%; 109/139) (**Table 3**). The presence of peridomiciles increased the risk of infestations by 12.03 times (p = 0.005) (**Table 3**). Of the total chicken coops, 64.2% were classified as vulnerable to colonization, with 10.28 (p = 0.005) times greater risk of being infested by triatomines than those that were not vulnerable (**Table 3**).

The distances between 73 houses and chicken coops were measured, with 57.5% (42/73) having a distance less than 12 m, of which 50% were infested with triatomines. There was no association between the distance from the chicken coops to the house and the infested houses.

# Knowledge and practices associated with the presence of triatomines

We interviewed 95 people who had an average age of 49 years (73.9% women and 26.1% men), of which 63.2% related the bugs to the transmission of the disease, but only 26.7% knew that transmission occurs through insect feces. Of all interviewed people, 35.8% (34/95) claimed to have seen bugs in the previous year. The houses and/or peridomiciles of 44.1% of these people (15/34) were positive for triatomine presence. Only 40% of these houses were sprayed. Of the people who reported having found bugs, 82% killed them and only 25% reported the infestation to local authorities.

Regarding practices preventing the presence of triatomines, 69.5% (66/95) of the interviewed people mentioned spraying with insecticides as the most commonly used measure. Only 10.5% of the respondents recognized the importance of keeping the peridomiciliary structures tidy and clean to avoid an infestation of triatomines. Of these people, 70% did not have their peridomiciles infested. A higher proportion of respondents (80%), who did not recognize the importance of the peridomiciliary for triatomine control, had infested peridomiciliary structures (**Table 4**).

The greatest lack of knowledge was related to congenital transmission and the possibility of treatment and cure. Only 12 people (11 of them females) recognized the congenital pathway.

TABLE 2: List of knowledge and practices included in the residents ' interviews.

- Heard about Chagas.
- 2. Recognize triatomines.
- 3. Know that bugs transmit Chagas.
- 4. Know about congenital transmission.
- 5. Know about transfusional transmission.
- 6. Recognize the importance of improving housing.
- 7. Know about the control of triatomines by spraying the house with insecticides.
- 8. Recognize the importance of cleaning the peridomiciles.
- 9. Consider important that animals sleep outside the house.
- 10. Recognize that Chagas can be cured and treated.

Regarding knowledge about whether Chagas disease can be cured, 38.9% answered positively, although they had doubts about who was cured. Most respondents answered that some people could be cured, and only 16% indicated that children are most likely to overcome the disease (**Table 4**).

Among the interviewed people, 24% had Chagas disease. However, 13% did not know if they had Chagas as they had never had a diagnosis. Most people claimed to have heard about Chagas disease. However, 63% of these people did not know about any treatment, 86.5% did not know about congenital transmission, and 93.3% were unaware of the transfusion route. The main source of information about this disease were from Chagas technicians (36%) and health care personnel (22%).

### **DISCUSSION**

The results in this study indicate the persistence of *T. infestans* populations in rural areas of northwestern Córdoba. The low percentage of domiciliary infestations found in this work is probably not only due to a few vulnerable houses recorded in the study area, but also due to the low sensitivity of the manual method used for searching for triatomines (hour-man technique). Previous investigations of routine surveillance systems used in the context of detecting vector-borne diseases like Chagas, have shown that these systems have sensitivity estimates frequently below 50%, likely resulting in underestimates in the real infestation vector numbers<sup>36,37</sup>. Historically, the house has been considered a risk factor, especially those houses whose structure offers suitable refuge for triatomines.

The type of construction material as a risk factor has been observed by several authors<sup>31,32,33</sup>, with degrees of risk depending on the environmental and social context, plus the region and

**TABLE 3:** Binary analysis of house unit type, structural risk factors, and residents' practices associated with the presence of triatomines in the domicile and peridomicile in rural communities of Cruz del Eje, Córdoba province, Argentina.

Structural risk factors in households and peridomiciles	Percentages (n)	Odds ratio Infested domiciles (CI 95)	Odds ratio Infested Peridomiciles (CI 95)
Both unplastered concrete or brick roof or thatched roof), and unplastered walls	26 (27)	13.21 (1.40-124.20)	-
Only walls	19.2 (20)	20.75 (1.17-197.9)	-
Only roofs	15.9 (20)	7.23 (1.12-46.65)	-
Chicken coops	78.4 (109)	-	12.03 (3.81-37.99)
Colonizable chicken coops	64.2 (70)	-	10.28 (1.75-60.4)
Distance < 12 m from chicken coops to houses	57.5 (42)	-	-
Risk factors related to the settlers' lifestyle			
Untidiness inside house	71.6 (47)	-	-
Animals sleeping inside the house	1.8 (1)	-	-

CI95: 95% confidence intervals.

**TABLE 4:** Knowledge of the population about Chagas disease, its vectors, and preventive measures in rural communities of Cruz del Eje, Córdoba province, Argentina.

Ítems	Percentage of affirmative answers out of a total of 95 respondents (n)	
1. People who heard about Chagas	92.6 (88)	
2. People who recognize triatomines	91.6 (87)	
3. People who know that bugs transmit Chagas	63.2 (60)	
4. People who know about congenital transmission	12.6 (12)	
5. People who know about transfusional transmission	2.1 (2)	
6. People who accept the importance of improving housing conditions	44.2 (42)	
7. People who know about the spraying of the house with insecticides for the control of triatomines	69.5 (66)	
8. People who accept the importance of maintaining the peridomiciles clean	10.5 (10)	
9. People who consider the importance of animals sleeping outside the house	56.8 (54)	
10. People who accept that Chagas disease can be cured and treated	38.9 (37)	

the type of vector<sup>21</sup>. In this work, although both roofs and walls associated with risk factors (unplastered, or thatched roofs with unplastered walls) were significantly associated with the presence of triatomines, the unplastered walls were associated with a greater risk. Other authors found that the rate of infestation in plastered houses is lower than in unplastered ones<sup>34,35</sup>.

The low percentage of domiciliary infestation found in this work is probably due to few vulnerable houses being recorded. Most of the houses with adobe walls and thatched roofs were

replaced through the "Programa de erradicación y sustitución de viviendas rancho <sup>38</sup> In contrast, there was a high infestation of triatomines in the peridomiciles, mainly associated with the presence of chicken coops, which are the most frequent peridomiciliary annex in the area.

The availability of hosts and shelters for triatomines in the peridomiciles is diverse and depends on the regions and the activities conducted inside those structures. In areas of the Humid Chaco, substantial differences were found in relation to the availability of hosts and local characteristics of habitat suitable for triatomine when compared to other areas of the Dry Chaco in Argentina<sup>19,22,38</sup>. Goat and pig corrals infested with *T. infestans* were identified as the main habitats and sources of re-infestation by insects in the Dry Chaco of Argentina<sup>39,40,41</sup>.

Alternatively, the materials used for the construction of corrals or chicken coops are also important, as different materials provide various types of refuge for triatomines. In the department of Ayacucho (San Luis) Chartier & Crocco<sup>13</sup> found that stone pile corrals appear to be the most suitable refuge sites for triatomines, probably due to the large number of empty spaces characteristic of this type of construction, and the great difficulty associated with using chemical control methods. In this work, the goat corrals were not frequent annexes and were rarely infested, probably due to the type of construction (stick palisade wall) offering few refuge sites for insects.

The analysis of factors related to the infestation by triatomines indicated that peridomiciles and, in this case, chicken coops, continue to be a challenge for control activities. Chicken coops were the most important annexes that were infested with triatomines, which shows a significant association between these types of annexes and infestations. Chicken coops built with sticks, wood, or cardboard (hen houses) were the most vulnerable because they provide excellent refuge sites for triatomines, which is in agreement with previous findings<sup>14,22</sup>. Preliminary results (C Soria: unpublished data) from a food profile within the same area indicate that most *T. infestans* collected in the peridomicile fed exclusively on chicken blood. However, the presence of combined blood sources was higher than expected, with blood meals being acquired from four hosts (dog, goat, chicken and human).

The persistence of triatomine in the peridomiciles within the study area is possibly related to the features of the peridomiciliary annex structures, especially of the hen houses. In these annexes, unlike goat pens, the presence of hosts is permanent, as chickens are bred year-round. In addition, these annexes offer optimal refuge sites that prevent good penetration of insecticides. Numerous works point out the importance of the peridomiciles in the maintenance of triatomine colonies and in the re-infestation processes, as well as the difficulty of spraying them<sup>18,42,43</sup>. Hen houses in particular are considered a suitable environment for the development of triatomines, where insect colonies contain numerous individuals, have a good nutritional status, and contain highly fertile females<sup>17</sup>.

The daily activities of the settlers and their lack of knowledge about the management of peridomiciles complicates the control of triatomine populations. Settlers ignore the fact that peridomiciles are important for maintaining triatomine populations and only consider the house as a risk factor where spraying is the only control method. Although they recognize that bugs can settle in the chicken coops, they do not know which actions can be taken to prevent, or reduce, the risk of infestation of these insects in these habitats and eventually in their homes.

In summary, people know where triatomines develop and that they must improve the quality of the houses and their peridomiciles. However, they do not take specific control actions, nor do they perceive the risks involved when not taking any control actions. This suggests that information alone is not enough. We need to provide these communities with tools and strategies, which are based upon their own perceptions, to solve these problems, and possibilities exist that the people will have to modify their environment.

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