Household risk factors associated to infestation of *Triatoma dimidiata*, the Chagas disease vector in Central Region of Veracruz, Mexico

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**Sandoval-Ruiz CA, Guevara R, Ibáñez-Bernal S.
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Salud Publica Mex 2014;56:213-220.**

Abstract

Objective. To evaluate risk factors facilitating the colonization of dwellings by *Triatoma dimidiata* in the central region of the state of Veracruz. **Materials and methods**. We applied socioeconomic questionnaires and entomologic surveys in three localities (Chavarrillo, Soyacuautla and Arroyo Agrio) in central Veracruz involving 115 households. **Results**. We found that the main risk factors were the predominance of unplastered walls and particularly those made of light weight aggregate concrete blocks and wood. At Chavarrillo, houses usually have unplastered walls, whereas in Soyocuautla walls are commonly manufactured with wood. In Arroyo Agrio, the phenomenon was seasonal, and bugs were commonly found in the dry season, particularly in relatively new houses, less than 20 years old. **Conclusions**. These results help to improve the surveillance capacity for this vector and the control strategies to reduce the transmission of Chagas disease in the state of Veracruz and other sites where this species is present.

Key words: *Triatominae*; risk factors; vector control; Chagas disease; Mexico

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Factores de riesgo intradomiciliarios para la infestación de *Triatoma dimidiata*, transmisor de la enfermedad de Chagas, en la región central del estado de Veracruz, México.
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Resumen

Objetivo. Determinar los factores de riesgo que facilitan la colonización intradomiciliaria de *Triatoma dimidiata* en la región central del estado de Veracruz. **Material y métodos**. Se aplicaron encuestas socioeconómicas y entomológicas en 115 casas en tres localidades (Chavarrillo, Soyacuautla y Arroyo Agrio). **Resultados**. El principal factor de riesgo para la colonización intradomiciliaria de *T. dimidiata* fue la presencia de paredes sin revocar, especialmente aquellas construidas con block y madera. En Chavarrillo el factor principal fueron las paredes sin revocar, en Soyacuautla las paredes de madera y en Arroyo Agrio las casas con menos de 20 años de haber sido construidas, junto con la temporada de secas. **Conclusión**. Los resultados encontrados pueden coadyuvar a mejorar los programas de vigilancia y control entomológico con el fin de reducir la transmisión de la enfermedad de Chagas vía vectorial en el estado de Veracruz y otros estados donde *T. dimidiata* puede estar presente.

Palabras clave: *Triatominae*; factores de riesgo; control de vectores; enfermedad de Chagas; México

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Received on: May 13, 2013 • **Accepted on:** October 30, 2013
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Chagas disease or American trypanosomiasis is one of the most important zoonotic diseases in the Americas with about 28 million persons carrying the protozoan. *Trypanosoma cruzi* (Chagas, 1909) infection in humans is gained by contact with feces of infected bugs.[1] In Mexico between one and three million people are infected with *T. cruzi*, and about 30 million are at risk of being infected.[2] It is well known that triatomine bugs are widely distribute in all tropical regions of Mexico, and infection prevalence occurs across the country, yet very little has been done to gain insights into how to handle the transmission of the disease.[3]

In the state of Veracruz it has been estimated that Chagas disease prevalence is between 2.8-3.0%,[4] representing 23% of cases in Mexico for the last ten years (Boletín Epidemiológico Nacional, 2002-2011). A total of ten triatomine species have been reported in Veracruz.[5] Most of them present low-abundance populations and are mostly confined to natural habitats, thus having little contact with human dwellings, exception made of *Triatoma dimidiata* (Latreille, 1811).[5]

In Veracruz, *T. dimidiata* has a wide range of hosts, and it is found in a diverse array of natural environments, anthropogenically modified rural habitats, and suburban areas, around and inside human dwellings.[5,6] Because of this, *T. dimidiata* is one of the most important species from the epidemiological point of view, not only in the state of Veracruz but also for the Yucatan Peninsula (Campeche, Quintana Roo and Yucatan states) and Central American countries.[7]

In general the highest number of Chagas disease cases in Latin America are found in rural areas where the domiciliary infestation by triatomine bugs is facilitated by the poor conditions of dwellings and by the deterioration of the landscapes which probably produce a reduction of natural wild hosts available for the triatomine.[8-16] Both conditions favored the bug contact with humans and therefore increasing the risk of infection with *T. cruzi*. For this reason, it is important to assess at local scale the risk factors related to the domiciliary infestation by *T. dimidiata*. This in turn will help to implement or improve the control strategies by targeting those aspects that most favor home infestation by this triatomine.[14]

In this work a detail characterization of dwellings (e.g. construction materials of floors, walls, and roofs) and inhabitant habits (e.g. rest of domestic animals inside houses) together with a systematic survey for bugs to evaluate potential factors facilitating the colonization of dwellings by *T. dimidiata* in three localities of the central region of the state of Veracruz.

Materials and methods

Study area

Three localities of the central region of Veracruz belonging to the Sanitary Jurisdiction V were assessed (figure 1):

Chavarrillo (19° 25´ 31″ N, 96° 47´ 29″ W), municipality of Emiliano Zapata. This locality is located at an altitude of 880 m with around 1 275 people. The climate is sub-warm and sub-humid with an annual mean temperature of 18ºC and annual mean precipitation of 2 779 mm.

Soyacuautla (19° 34´ 34″ N, 96° 34´ 40″ W), municipality of Actopan, is located at an elevation of 280 m and includes 710 people. The climate is warm and sub-humid with an annual mean temperature above 22ºC and abundant precipitation during summer and the beginning of autumn. Total annual rain is on average around 1 200 mm.

Arroyo Agrio (19° 42´ 20″ N, 96° 25´ 51″ W), municipality of Alto Lucero at an elevation of 20 m, and with a population of only around 100 people. The climate is warm sub-humid, annual mean temperature above 22ºC, abundant precipitation during summer and the beginning of autumn, with considerable less rain falling in winter. Total annual rain averages 1 192 mm.

In the three localities, the principal economic activities are agriculture and cattle ranching, but there is a high tendency of people to emigrate to nearby cities.[17] We applied socioeconomic questionnaires in three localities (Chavarrillo, Soyacuautla and Arroyo Agrio) involving 115 households over to two years sampling.

Triatomine sample and entomological indexes

Samples were collected over two years (February 2008-September 2009). Annual surveys at each locality were conducted every four months covering the three climatic seasons recognized in the region (dry, rainy, and *nortes* (cold fronts, gales). Based on the villages´ layouts, 1 ha^2 plot was randomly selected and surveys for triatomine were conducted in all houses within this area. We followed the methodology recommended by the Expert Committee on Chagas disease of World Health Organization,[18] and searched for triatomine in the interior of the residences, and walls of the houses to the edge of property generally bounded by a woof fence or stone wall. To facilitate the collection of bugs a commercial insecticide was applied to wall crevices,

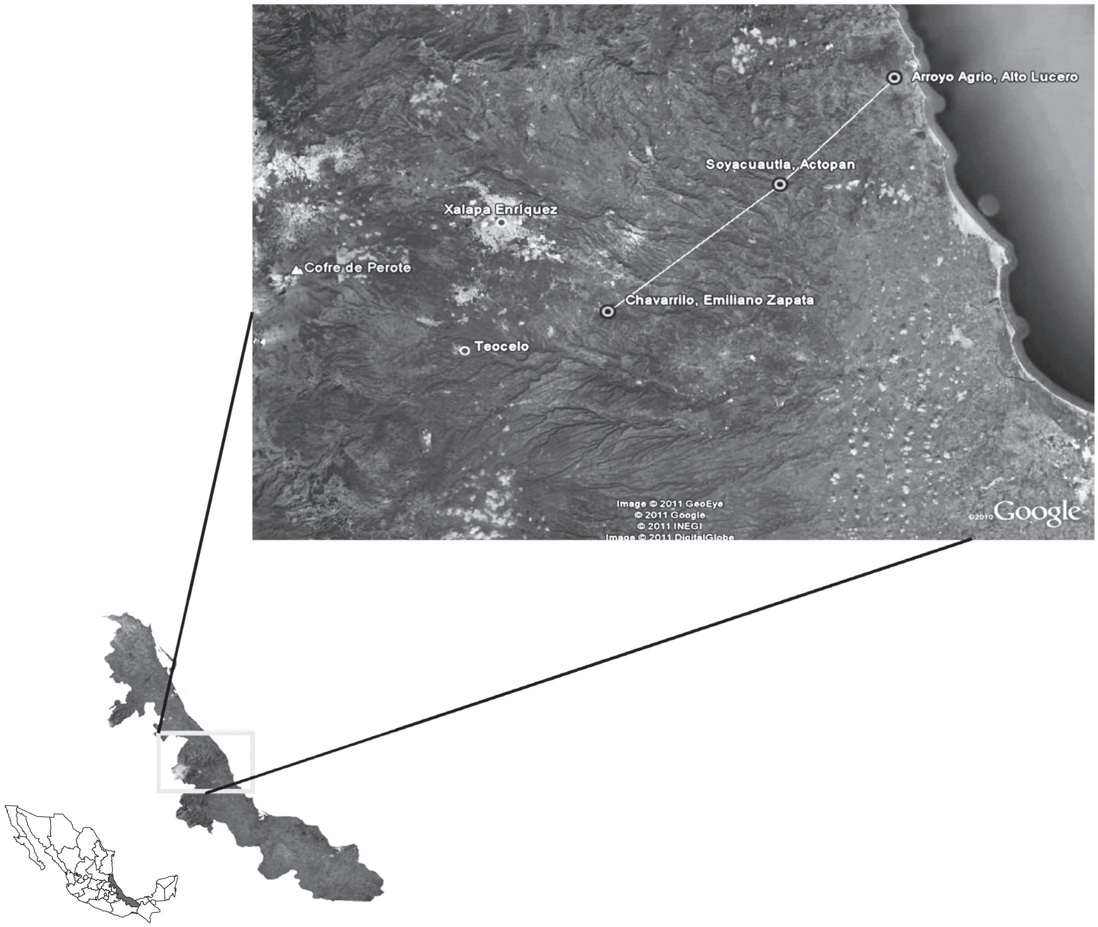

FIGURE 1. LOCALITIES SAMPLED IN CENTRAL VERACRUZ, MEXICO, FOR *T. DIMIDIATA* RISK FACTORS INFESTATION ANALYSIS (GOOGLE EARTH VER. 6.1.0.5001). 2008-2009

under the beds, to the roofs, woodpiles and other possible bug resting sites inside the houses and outside, fences of chicken coops, farmyards. This activity lasted 1 hour/man in each house.

Insects were collected and maintained alive in plastic container. For every collected bug a detailed description was made of the conditions were the insect was found. In the laboratory, samples of bugs feces were obtained by abdominal pressure, diluted in phosphate-buffered saline (PBS) and examined with a compound microscope at 20X to detect *T. cruzi* flagellates. Triatominae were examined with a stereoscopic microscope for taxonomic identification using dicotomic key proposed by Lent and Wygodzonsky,[19] we considered the classification proposed by Galvão and collaborators.[20]

Infestation (number of houses with positive records of triatomines), crowding (triatomines/infested houses), colonization (percentage of infested houses where triatominae nymphs were found) and natural infection (percentage of triatominae infected with *T. cruzi*) entomological indexes were obtained following the recommendations by WHO[18] and the Mexican Official Norm 032.[21] Additionally, at every house investigated an interview with the owner was conducted to obtain additional information on the house characteristics and inhabitant costumes (table I).

Statistical analysis

The bug-presence risk factors analysis were made in two steps: first a descriptive analysis by means of the technique of forest of classification tree models[22,23] to explore the relationship between variables obtained during the sampling with the presence of triatomine bugs

Table I

Summary of variables applied to survey localities in Central Veracruz, Mexico, as possible risk factors associated to *T. dimidiata* infestation inside household. 2008-2009

Variables		
Altitude	Wall construction materials	Animals sleeping inside house
Localities	Plaste ron walls	Places where animals sleep inside house
Number of persons that live in house	Roof materials	Chicken cops at peridomicile
House age	Floor materials	Rock piles at peridomicilies
House with rooms	Use of insecticides intradomestically	Firewood at peridomicilies
Number of bedrooms	Firewood inside house	Backyard garden
Number of beds	Agriculture material inside house	Vegetation type around house
Number of person in each room	Building materials inside house	Wild animals in ground house
Knowledge of triatominae by householders	Domestic animals at peridomicily	Season of the year

(table I), using the statistic package R* Analysis routine consisted in the use of function "forest of trees", set to sample randomly 1 000 times 80% of observed data and estimated a classification tree for each sample to then reconstruct the consensus classification tree.

The second step was analytic, using a generalized lineal model (GLM) with a binomial error structure and the link function "logit". The response variable was the presence/absence of triatomine bugs at each revised house. Explanatory variables for the initial model were selected according to the results of the forest of classification tree models. Following the initial adjustment a model simplification protocol was followed based on ANOVA test between models.[22] Both set of analysis, classification trees and GLM were done for the overall data as well as separately for each of the three localities to detect general and particular patterns related to the presence of *T. dimidiata*.

* R Development Core Team. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. 2010. Available in: www.cran.r-project.org/

Results

Entomological indexes

The entomological indexes obtained are shown in table II. Natural infection with *T. cruzi* was different not only between localities but also between the studied years (2008 and 2009), being Soyacuautla the locality in which the variation was considerable. Colonization indexes were high in Chavarrillo and Soyacuautla (88 and 100% respectively), whereas in Arroyo Agrio nymphs of triatomine were collected only in 66% of houses. Infestation by *T. dimidiata* in Chavarrillo and Arroyo Agrio was different between the years of study, being highest in the second year. In Soyacuautla bug infestation was similar in both years. Crowding index was similar for Chavarrillo and Soyacuautla during the two years, but in Arroyo Agrio a difference was observed with an increment of nearly three times from 2008 to 2009 (table II).

Risk factors

With the general analysis of classification trees (figure 2A) we found that the binary factor "plaster wall/

Table II

Domiciliary entomological indexes for *T. dimidiata* at Central Veracruz, Mexico, during 2008-2009

	Infestation Index (%)		*Colonization Index (%)*		*Crowding Index (%)*		*T. cruzi infection Index (%)*		*Total number of triatominae collected*	
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009
Chavarrillo	20.8	42.8	80	100	9	9	17.4	11.6	45	54
Soyacuautla	18.5	16.6	100	100	6.8	9.6	14.3	4	34	29
Arroyo Agrio	21.4	44.4	66	62.5	2.3	6.8	0	2.7	7	55

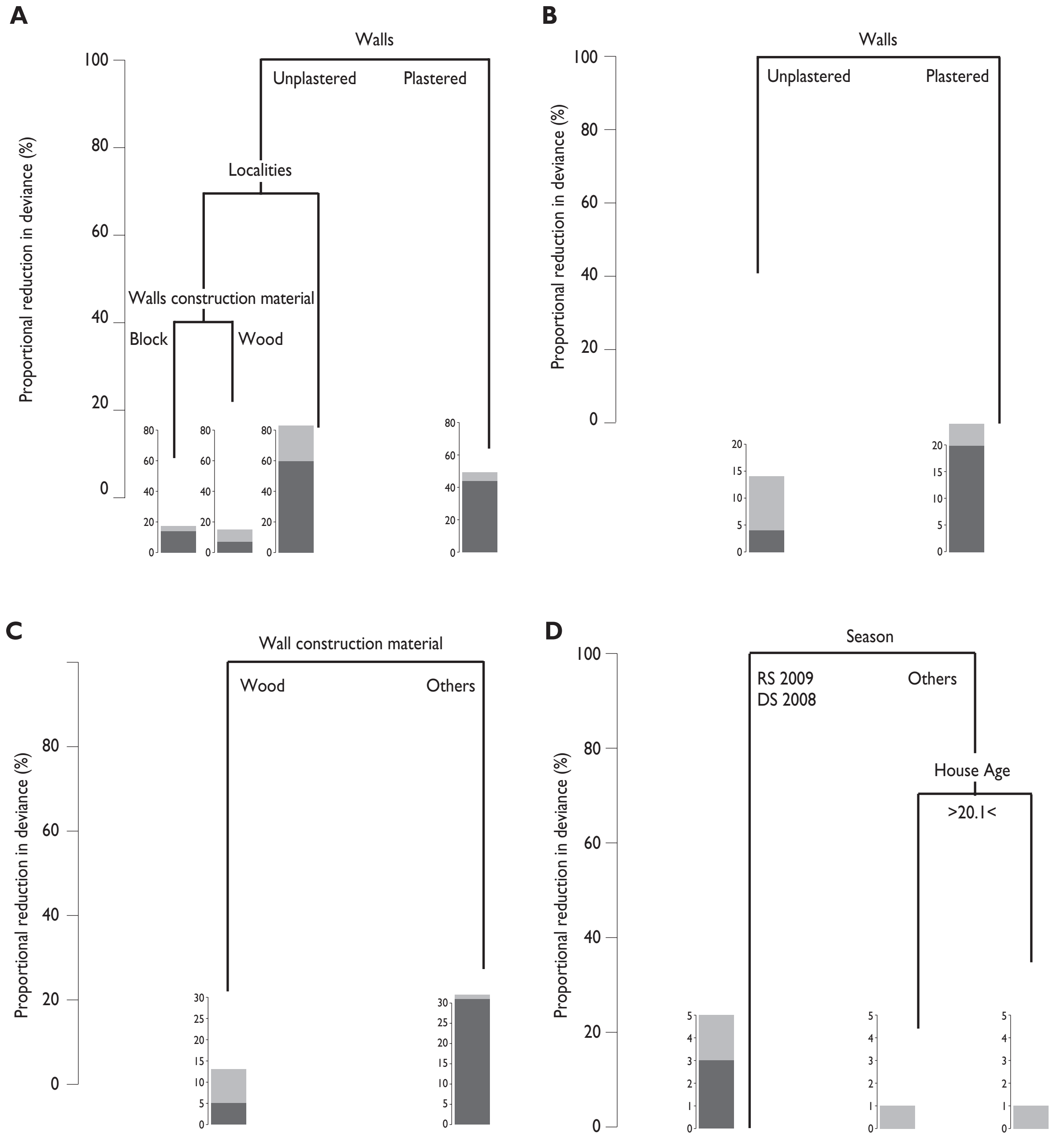

Figure 2. Classification tree analysis of domiciliary risk factors for *Triatoma dimidiata* infestation. A) The explanatory variables were walls (unplastered or plastered), localities (a= Arroyo, Agrio, b= Chavarrillo, c= Soyacuautla), and wall construction material (block or wood). B) Classification tree analysis for Chavarrillo (municipality of Emiliano Zapata). The explanatory variables were walls (unplastered or plastered). C) Classification tree analysis for Soyacuautla (municipality of Actopan). The explanatory variables were walls construction materials (wood or others). D) Classification tree analysis for Arroyo Agrio (municipality of Alto Lucero). The explanatory variables were: Season (RS= Rainy season – DS= Dry season or others) and house age (>20.1<). Histograms under the terminal nodes represent the numbers of observations (triatomine absence (dark gray) or present (light gray) inside houses) by each variable. Numbers separated by a slash represent absence/ present triatomine.

unplastered wall" was the variable that better explains the triatomine presence inside the house. From all the analyzed houses with unplastered walls 25% had *T. dimidiata*, whereas *T. dimidiata* was found only 4% of houses with plaster walls. Following the descendant order of the classification tree, *T. dimidiata* was most common in houses without plaster walls of Arroyo Agrio and Soyacuautla (16.5%), as compared with Chavarrillo (8.9%), especially when the walls were made with block or wood (13.9%) in comparison with houses with walls constructed with bricks or metal sheet (0.8%).

Analyzing by localities, it was observed that Chavarrillo had most houses with unplastered walls and consequently most infested by triatomine bugs (figure 2B), in Soyacuautla houses with walls built with wood were the principal risk factors for the presence of *T. dimidiata*, being safer those houses with walls of block, metal sheet or bricks (figure 2C). For Arroyo Agrio it was not possible to detect intrinsic characteristics of the house related to the presence of triatomine bugs. For this last case, the analysis showed that the season was the only significant factor associated with the presence of *T. dimidiata* (figure 2D); in the dry season of 2008 (DS 2008) and in rainy season of 2009 (RS 2009).

The global analysis of GLM, as the classification tree, showed that wall materials, unplastered walls and the locality were the main factors that explain the intradomiciliary bug presence (table III). Additionally, the interaction between localities and wall materials (different to block walls) (χ^2=4.95, gl=106, p=0.063) were marginally significant and thus a potential risk factor (table III). The GLM for Chavarrillo showed that houses with unplastered walls (χ^2=11.63, gl=36, p=0.001) is the principal risk factor for the intradomiciliary presence of *T. dimidiata*; for Soyacuautla the principal risk factor was the wood walls (χ^2=18.81, gl=43, p=0.0001), and for Arroyo Agrio the interaction between season and the age of the house (<20 years old represents the principal risk factor for this locality) (χ^2=14.35, gl=43, p=0.001). Houses with less than 20 years old were more susceptible to be infested with triatomine bugs in dry seasons as compared with older houses in the rainy season.

Discussion

In the present study was observed that unplastered walls (independently of the statistic exploratory analysis by classification trees or analytic models (GLM)) were the principal risk factor for the intradomiciliary presence of *T. dimidiata*. This is consistent with previous reports of *T. dimidiata* in Costa Rica and Guatemala,[10,15] and *T. infestans* (Klug 1834) in northern Argentina.[8]

Unplastered walls are a good refuge substratum for tiatomine adults and nymphs due to the numerous holes and crevices that serve as resting sites for the bugs. In a previous study conducted in Veracruz State, Mexico, Salazar[12] mentioned roofs made of palm leaves were the principal risk factor for intradomiciliary colonization of *T. dimidiata*, but this does not agree with our results. It is likely that Salazar´s[12] finding comes from a bias interpretation of data. Houses roofed with palm leaves are common in rural areas of Veracruz State examined, as some houses in the area present palm roofs along the geographic distribution but *T. dimidiata*, is very rarely found at the roof level, this species is most common below 1.50 meters, near the floor.[24, 25]

Table III

Risk factors analysis using generalized lineal model (GLM) considering three localities in altitudinal gradient at Central Veracruz, Mexico, during 2008-2009

	Df	*Deviance*	*Resid. Df*	*Resid. Dev*	*P(>|Chi|)*
NULL	NA	NA	114	139.641	NA
Unplastered walls	1	16.822	113	122.819	4.104E-05*
Localities	2	8.269	111	114.550	0.016§
Wall construction materials (no block)	1	6.758	110	107.792	0.009‡
Unplastered walls:Localities	2	3.048	108	104.744	NS
Localities:Wall construction materials (no block)	2	4.957	106	99.787	0.063

Significancy codes:
* 0
‡ 0.001
§ 0.05

NS: nonsignificant value
NA: no apply

Other consistent variable that can be considered as a risk factor is the wall material (wood, adobe), which has a direct relationship with the availability of refuges in unplastered walls, as it has been observed in Colombia, Argentina, Brazil, Ecuador, Guatemala and some states of Mexico (as Morelos and Estado de México) for bugs species such as *Rhodnius prolixus* (Stål 1859), *R. pallecens* (Barber 1932), *Panstrongylus geniculatus* (Latreille 1811), *Triatoma maculata* (Ericsson 1848), *T. dimidiata*, *Meccus pallidipennis* (Stål 1872) specially at nymph stage.[8-11,13-16,26]

We founded that dirt floor do not represent the principal risk factor for triatomine bugs infestation like in Costa Rica or Guatemala.[10,15] In this study only 22.6% of surveyed houses have this condition and 42% of them were recorded with intradomiciliary triatomine. This could be a consequence of the Mexican government program named "Piso firme" (concrete floor) that have been applied during the ten last years with the aim to improve residents quality of life, reducing respiratory, gastrointestinal and dermic illness of the rural people. Indirectly it contributes to limit the triatomine infestation of houses, as had been documented in Costa Rica.[16,27]

Risk factors are not present as isolated variables, and the presence of more than one apparently increase the possibility of triatomine infestation. The GLM analysis showed that variables in localities as Chavarrillo and Arroyo Agrio in which walls are made with some materials but rarely with block promote the highest infestation indexes (table II).

It has been demonstrated that risk factors related to triatomine infestation can be different in different regions[11] even if it is the same vector species (e.g. *T. dimidiata*), which can be explained by differences in climate conditions, soil use and human costumes. In Chavarrillo the presence of houses with unplastered walls is correlated with highest infestation and infection indexes. In Soyacuautla houses with wood-walls had a colonization index of 100% during the years of study, and in Arroyo Agrio the dry season probably is correlated with the flight of wild triatomine and the attractiveness of houses light bulbs as it was demonstrated in Yucatan.[28] The age of the house (less than 20 years old) is another important risk factor as was mentioned in a study in Morelos,[13] probably due to the abundance of holes and crevices in the walls.

It is important to mention that only in 33.9% of the sampled houses, people use commercial insecticides against insects (mosquitoes), bugs infestation were lower compared with houses were doesn´t use insecticides. We think that people doesn´t have a correct handling of insecticides. This fact together with precarious conditions of houses could be an additional factor related to the highest infestation, crowning and colonization indexes obtained in the collection sites.

The main variables identified in this work will help to improve the integral control procedures against triatomine bugs, considering not only the use of chemical insecticides at the beginning of the campaign but also the improvement of houses´ walls and floors, considering that the proposed changes do not alter their cultural patterns and they do not represent an extraordinary investment in their properties,[14] to prevent the subsequent colonization by triatomine bugs. This strategy has been implemented in Central American countries like Guatemala[29] were the synergy among sanitary authorities and people has showed that infestation of houses by triatomine bugs diminished and consequently transmission of *T. cruzi*.

Acknowledgements

This study was supported by Fomix-Conacyt – Gobierno del Estado de Veracruz under the Project: "Estratificación de Tres Enfermedades Prioritarias en Zonas de Riesgo con Base en sus Insectos Vectores en el Estado de Veracruz" [No. 68317] granted to SIB, and was additionally supported by the Scientific Collaborative Agreement between the Ministry of Health of the State of Veracruz (Sesver) and Instituto de Ecología, AC (Inecol). CASR was supported by scholarship from the Mexican Council of Science and Technology [Conacyt, No. 165670]. We appreciate the help of Fredy Mendoza, Ruth A. Hernández Xoliot and technical staff from the Sesver (Ministry of Health of the State of Veracruz) Entomology Department from Sanitary Jurisdition V, María Teresa Suárez Landa, Antonio Abella Medrano, Adriana Beltrán Aguilar, Ana Lilia Gutiérrez from Inecol.

Declaration of conflict of interests. The authors declare that they have no conflict of interests.

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