

Artículo Original**MORPHOMETRIC DISTINCTION OF DOMESTIC AND SYLVATIC POPULATIONS OF *Rhodnius ecuadoriensis* FROM DIFFERENT GEOGRAPHICAL ORIGINS****DISTINCIÓN MORFOMÉTRICA DE POBLACIONES DOMÉSTICAS Y SILVESTRES DE *Rhodnius ecuadoriensis* DE DIFERENTES ORIGENES GEOGRÁFICOS****JS. Patterson+ F. Abad- Franch +, CA. Cuba Cuba++ & MA. Miles+**

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Recibido: 5 de abril, 2020. Aceptado: 19 de mayo, 2020

ABSTRACT

Rhodnius ecuadoriensis (Hemiptera, Reduviidae) is an important vector of both *Trypanosoma* hemoflagellates, *Trypanosoma cruzi* and *Trypanosoma rangeli*, in Ecuador and Peru. Ecotopes of sylvatic and domestic/peridomestic habitats have been reported in Ecuador. Meanwhile in Peru, to the best of our knowledge, findings of sylvatic populations in their different ecosystem regions have not yet been documented. Could this be the product of a lack of appropriate studies on wild populations of triatominae in Peruvian environments? In order to elucidate this topic, we take advantage of new insights in geometric morphometry as a tool to help differentiate between wild populations and the corresponding domestic/peridomestic ones, collected in their respective environments. When analyzing our results, we confirmed the efficacy of this technique in our study, and furthermore, we believe that it could be a proper tool for rangelirosis and Chagas disease vector control surveillance in Ecuador and Peru.

Keywords: Morphometric geometry, sylvatic, domestic/peridomestic populations distinction, *Rhodnius ecuadoriensis*.

RESUMEN

Rhodnius ecuadoriensis (Hemiptera, Reduviidae) es un importante vector de los tripanosomas *Trypanosoma cruzi* y *Trypanosoma rangeli* en el Ecuador y Perú. Se han reportado ecotopos de hábitats silvestres y domésticos/peridomésticos en Ecuador. Sin embargo, en Perú, hasta donde sabemos, no se ha documentado hallazgos de dichas poblaciones silvestres. ¿Podría este ser el caso de una falta de estudios focalizados en la búsqueda de poblaciones silvestres de triatominos dentro de los diferentes ecosistemas del Perú? Para elucidar este tema, aplicamos nuevas perspectivas en morfometría geométrica, como una herramienta que podría auxiliar en la diferenciación de poblaciones silvestres de aquellas domésticas/peridomésticas, colectadas en sus respectivos ambientes naturales. Al analizar nuestros resultados, se confirmó la utilidad de esta técnica dentro de nuestro estudio, y esto nos llevó a creer asimismo que serviría como un elemento apropiado en el control vectorial de la enfermedad de Chagas y de la rangelirosis, en Ecuador y Perú.

Palabras claves: Geometría morfométrica, silvestre, diferenciación de poblaciones domesticas/peridomesticas, *Rhodnius ecuadoriensis*.

1. INTRODUCTION

Rhodnius ecuadoriensis (Lent y Leon, 1958) is a significant vector of Chagas disease in the Western side of the Andes of Central - Southern Ecuador. In Peru, this insect is the vector of *Trypanosoma rangeli*, even though its role on the epidemiology of Trypanosomiasis americana has not yet been defined (Cuba Cuba et al., 2002). Sylvatic populations breed in *Phytelephas aequatorialis* palm trees in subtropical valleys of Pichincha, Ecuador (Abad Franch et al., 2001). *R. ecuadoriensis* has a natural preference for a dry and xerophytic ecosystem, which is commonly seen in Northern Perú where no palms species are found. The species seems to be exclusively associated with human environments in southern Ecuador and over its entire range in Peru. In a preliminary work, some phenetic differences were recorded between sylvatic and domestic/peridomestic specimens (Abad-Franch, 2000; Patterson et al., 2002). Here we characterize those differences using multivariate statistics of morphometric variable, a technique that is able to discriminate among conspecific bug populations of sylvatic and domestic origin (Dujardin et al., 1997).

2. MATERIAL AND METHODS

A total of 65 adult *R. ecuadoriensis* (classified after Lent y Wygodzinsky, 1979) from four populations were studied: Chicama, Loja/Suyo, El Oro and Manabi (Table 1, and Figures 1 and 2). *Rhodnius pallescens* (15 individuals) was used as an outgroup (here called *pallescens*). Bugs from Peru were collected by Cuba (Cuba et al., 2003), in the Cascas valley, Chicama (Figures 2A, 2B).

Seven parameters were considered when measuring the head of each bug (Fig 3, 1 and 3, 2). Data was obtained by using a digital video camera mounted on a light dissection microscope and analysed by isometry-free canonical variate analysis (CVA). Log- transformed datasets were centred by row and submitted to principal component analysis, the first six principal components (PC) were used as input for canonical variate analysis. The first two canonical factors (CV 1 and CV 2) were subsequently used to construct a factorial map. Mahalanobis distances were submitted to an Unweighted Pair Group Method with Arithmetic mean (UPGMA) cluster analysis. A dendrogram was constructed showing the relationships between populations. The JMP and STATA 7 software packages were utilized. Size differences were analyzed by ANOVA, using the mean values of all log -transformed measurements of each bug ("isometric estimator of size").

3. RESULTS

Factorial maps derived from CVA (Figures 4 and 5) detected important differences between the sylvatic population of *R. ecuadoriensis* and those of peridomestic and/or domestic origins. There was also little overlap between domestic/peridomestic bugs from Ecuador and domestic Peruvian specimens from Cascas valley, Chicama (Table 1). However, these three groups showed limited variability among them. The UPGMA dendrogram (Figure 6) also reflects the separation between domestic and sylvatic *R. ecuadoriensis* populations.

The fact that the Chicama group is clustered together with El Oro is probably related to similarities in size (see Figure 7). In fact, the contribution of size (allometric residuals) to CV was important when examined by regression analysis. No differences were recorded between field-collected bugs from Manabi and those from the laboratory colony. Bugs of sylvatic origin were significantly bigger ($p < 0.05$) than synanthropic ones. (Figure 7). In addition, bugs from Loja-Suyo were smaller than those from Chicama and El Oro, which were very similar in size.

Table 1. Populations used in this study

GROUP	ORIGIN	Nº	HABITAT
Chicama	Cascas, Valley, La Libertad, Peru 7°28'S 78°49' W 1626 msol	14	Domestic
Loja	Lucero, Loja, Ecuador 4°15'S79°30'W1500 msol	15	Domestic / Peridomestic
Suyo*	Piura, Perú 4°30 S 80°51'11 msol	1	Domestic / Peridomestic
El Oro	Lourdes, El Oro, Ecuador** 3°40'S 79°39'W876 msol	18	Domestic / Peridomestic
Manabi	Portoviejo, Manabi, Ecuador 1°6'S 80°24'W130 msol	17	Palm Trees
<i>Rhodnius pallescens</i>	Laboratory colony	15	Colombia

*(See Discussion and Fig 5).

**Some specimens from a colony (Lab. NIRT FIOCRUZ-Rio de Janeiro, Brazil established in 1992 with bugs collected in the same Ecuadorian locality were also used)

4. CONCLUSIONS AND DISCUSSION

1. Utility of morphometric variables

Isometry-free morphometric analysis can establish clear-cut differences between sylvatic and synanthropic populations of *R. ecuadoriensis*. Thus, metric variables could be used to monitor the origin of re-infestations after residual spraying and it may give clues to explain the domiciliation process of the species. As pointed out, sylvatic triatominae constitute a challenge in vector control transmission (Guhl et al., 2009), so viable control strategies must emphasize the presence of bugs populations in wild ecotopes.

Differences between the Manabi population (sylvatic) and the El Oro-Loja Peru cluster (domestic) were easily detected using this approach. The sensitivity of the morphometric analysis was further demonstrated when a single bug distorted the Cascas, Chicama cluster (Figure 5). When the origin of bugs was checked, it was discovered that this single specimen had actually been collected from Suyo, a Peruvian locality a few km away from the site of capture of the Loja specimen (Lucero, see Figure 2), to which cluster it was assigned by the morphometric analysis. That group was therefore renamed (Loja-Suyo) and the Suyo specimen included in it. However, the largest part of the intraspecific variation detected does not seem to be geographic in nature. The Cascas, Chicama group (Table 1) bugs are most similar to those of El Oro, a distant locality. In general, all the synanthropic populations are similar to each other, and all of them are clearly different from the sylvatic one. These differences are therefore likely to reflect the distinct habitat.

Our results suggest that a baseline set of metric variables including different local populations could be used to investigate the origin of any re-infesting bug (see also Schofield, 2001). Therefore, the use of geometric morphometry could help overcome the influence of size in the analysis (Jaramillo, 2000).

2. Possible isolation of populations

The strong separation between Manabi and the rest of populations might also reflect a certain degree of genetic isolation. In fact, the palm trees that constitute the primary natural habitat do not exist in the arid north of Peru, nor in the Valley of Loja. Only in El Oro, scattered remnants of humid forests with *Phytelephas* palms can be found (cf. Abad-Franch et al, 2001). Together with the present results, this suggests that domestic and peridomestic populations of southern Ecuador and northern Peru (i.e. the epidemiological significant ones) are probably isolated from sylvatic foci associated with *Phytelephas* palms. Consequently, local eradication of those synanthropic populations may be attainable through residual insecticide spraying. Population genetic studies based on molecular approaches will contribute to clarify the relationships between these populations.

5. ACKNOWLEDGEMENTS

Supported by the ECLAT network. UNDP/ World Bank TDR (grant 970195), CAPES Ministerio de Educação e Cultura, Brasil), the Oficina Nacional de Epidemiología (Instituto Nacional de Salud. Ministerio de Salud Pública, Perú), the University General Hospital of Valencia, Spain and the Cañada-Blanch Foundation, Spain. F. Guhl and J. Jurberg kindly provided colony specimens from their laboratories.

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FIGURES

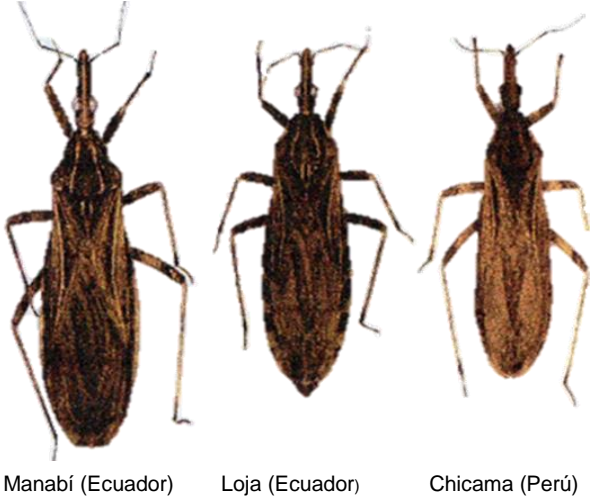


Figure 1. Different *Rhodnius ecuadoriensis* populations



Figure 2. Sites of capture (●) of *Rhodnius ecuadoriensis* populations



Figure 2A. Household infested with *Rhodnius ecuadoriensis* (Lent y León, 1958) - Adult female, below. Presence of eggs and molts inside of Gomez-Nuñez trap. Note the roof built with rice straw material. Cascas valley, Chicama, Peru.

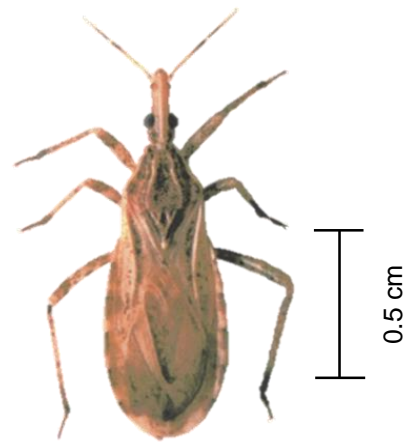
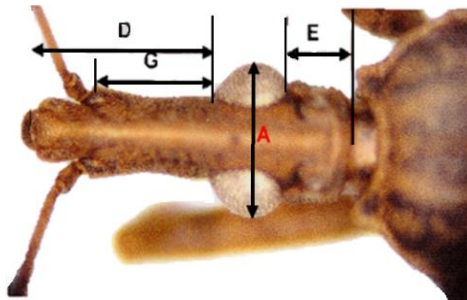
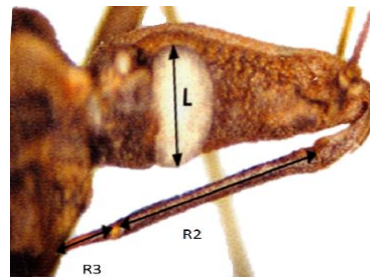


Figure 2B. *Rhodnius ecuadoriensis* individual.



3.1. Dorsal view

- A= Outer distance between eyes
- D= Antecular distance
- E= Postocular distance
- G= Length of antenniferous tubercle



3.2. Dorsal view

- L= Maximum diameter of the eye
- R2= Length of second rostral segment
- R3= Length of third rostral segment

Figure 3. Dorsal and lateral views of the head indicating morphometric measurements

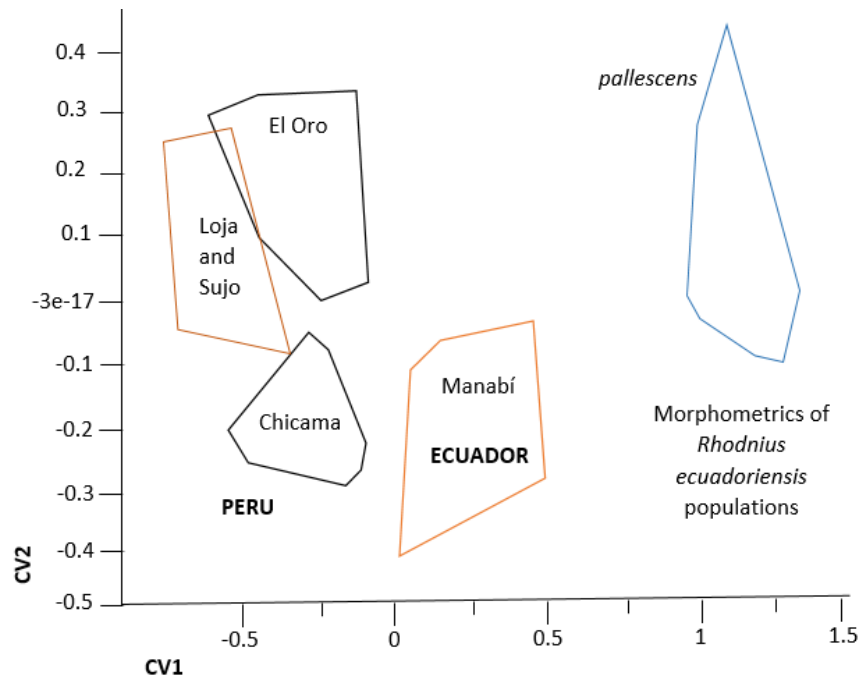
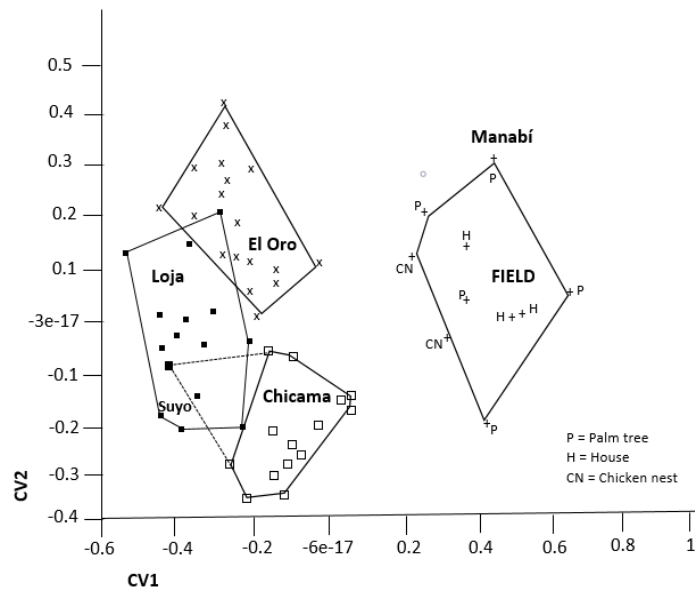


Figure 4. Isometry free - CVA



One *Rhodnius ecuadoriensis* from Peru (■) was closer to the Loja group than to other Peruvian specimens after CVA. The localities where this bug and those from Loja were collected (Suyo and Lucero, respectively) are less than 40 km away. No differences were recorded within the Manabí group, regardless of the origin of bugs.

Figure 5. Isometry-free CVA: Individual diagnosis of origin (without outgroup)

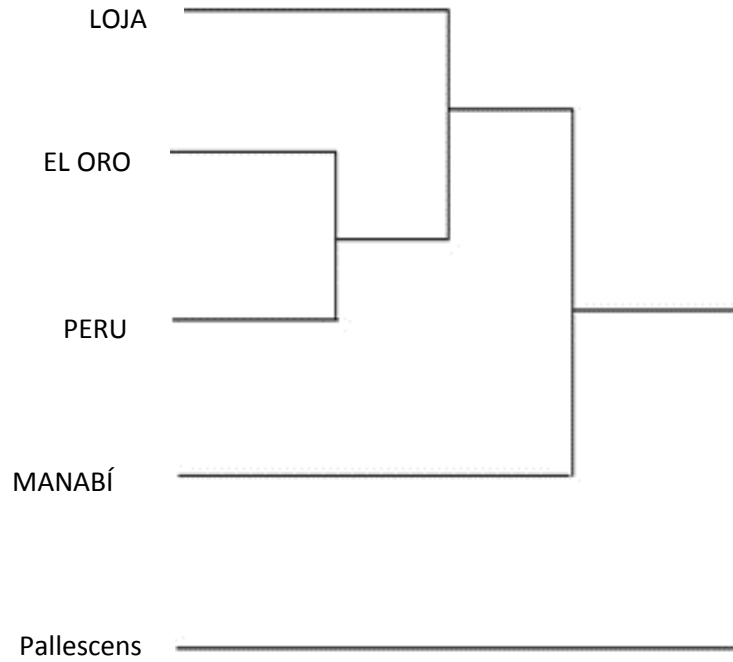


Figure 6. UPGMA dendrogram derived from Mahalanobis distances

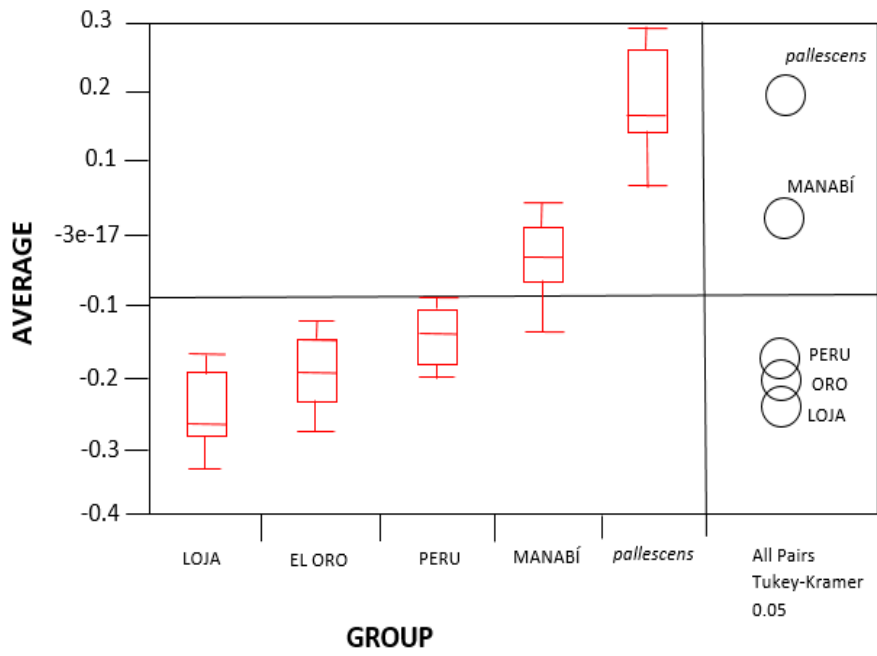


Figure 7. Oneway analysis of size (as the average of log-transformed measurements by individual).