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Differences on biological attributes of three populations of *Meccus pallidipennis* Stål (Hemiptera: Reduviidae)

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ABSTRACT

Background & objectives: Meccus pallidipennis is one of the most epidemiologically important vectors of *Trypanosoma cruzi* to reservoir hosts in nine states of Mexico. Triatomines occurring in distinct locations normally adapt to local conditions. The aim of this study was to examine the biological attributes of three populations of *M. pallidipennis* from areas with different environmental characteristics as a factor influencing the triatomine capacity for *T. cruzi* transmission.

Methods: The values of biological parameters related to the life cycle, the number of blood meals to molt to next instar, fecundity and percentage of females after a biological cycle of three populations of *M. pallidipennis* were evaluated. A cohort of each of the three studied populations from different geographical areas of Mexico was maintained under similar laboratory conditions and then compared with each other.

Results: The life cycle was less than six months in all the studied cohorts, with differences among them. The number of blood meals to molt was lower for the cohort from Izúcar de Matamoros. Laid eggs per day per female was lower for the cohort from Luvianos. In contrast, no important differences were recorded on the percentage of mortality, egg eclosion rate or percentages of obtained females.

Interpretation & conclusion: It was found that an important level of heterogeneity exist between the three studied populations of *M. pallidipennis*, apparently influenced by the remarkable differences on environmental conditions on the localities where the founders were initially collected, that emphasizes the necessity of studies on local populations of triatomines.

Key words Biological variation; environmental conditions; life statistics; *Meccus pallidipennis*; populations

INTRODUCTION

In Mexico, *Meccus pallidipennis* is considered as one of the most epidemiologically important vectors of *Trypanosoma cruzi* to human and animal reservoir hosts. It is distributed in nine states throughout western, eastern, central, and southern Mexico; it is the main vector of *T. cruzi* in six of them[1–3]. Triatomines occurring in distinct locations are normally adapted to local conditions[4]. Over time, ecological isolation and adaptation to local conditions may lead to the development of geographically isolated populations that differ in various biological traits, which gives rise to the variation in a range of population parameters[5–7]. Different populations of *M. pallidipennis* have been analyzed by using different techniques. The nucleotide sequences of the cytochrome B gene and the antennal phenotypes were analyzed for the specimens of the three populations of *M. pallidipennis* from different areas of Mexico. It was recorded that one of them was significantly far apart in a phylogenetic tree[8]. Complementarily, different biological traits have been studied and it has been concluded that they are important criteria to determine the relation between populations of related triatomines[9]. On this way, some studies[5–7] on different biological parameters (life cycle, feeding patterns, and fecundity of females) of *M. pallidipennis* and of *M. longipennis* (a close related species) have shown differences when populations from different geographic areas have been compared, providing an explanation to recorded differences on numbers of infected inhabitants on each of those areas.

To contribute to the explanation of the differences among populations of *M. pallidipennis* and, as a consequence, the differences on the potential role as vector of *T. cruzi* of each population, a study was carried out to investigate the life cycle and some related biological attributes of three populations of *M. pallidipennis* from three different environmental and geographical areas of Mexico.

MATERIAL & METHODS

Biological material (Triatomines)

The second generation from three laboratory colonies of *M. pallidipennis* recently established from at least 30 specimens collected from certain localities with different environmental characteristics in three different Mexican states (Fig. 1) were used; these localities were Luvianos, in the state of Estado de México; Chilpancingo de los Bravo, in the state of Guerrero; and Izúcar de Matamoros, in the state of Puebla. Luvianos (18° 55′ N, 100° 18′ W) is 1130 m above sea level; has a template climate and is characterized by the presence of pine (*Pinus* spp), holm oak and oak forest (*Quercus* spp), Mexican coniferous tree (*Taxodium mucronatum*) and Jacaranda (*Jacaranda* spp). Chilpancingo de los Bravo (17° 33′ N, 99° 30′ W) is 1647 m above sea level, has a sub-humid-template climate (temperatures ranging from 15°C to 24°C). The locality is mainly characterized by the presence of xerophila vegetation, like mezquite (*Prosopis* spp), huizache (*Acacia farnesiana*) and cazahuate (*Ipomoea arborescens*). Finally, Izúcar de Matamoros (18° 34′ N, 98° 33′ W) is 1300 m above sea level, has a calid-sub-humid climate and is mainly characterized by the presence of Bursera (*Bursera* spp), Brazilian stick (*Haematoxylum brasiletto*), Yuca (*Yuca* spp) and plenty of sown fields of sugarcane (*Saccharum officinarum*)[10]. The specimens were identified according to the taxonomic key of Lent and Wygodzinsky[11], taking into account the revalidation of the genus *Meccus*[12].

Triatomine maintenance

Although laboratory rearing imposes a certain degree of selection pressure on aspects of insect biology, all studied cohorts from recently colonized triatomines were exposed to the standardized environmental conditions that were favourable to their survival; hence, it was assumed that estimates of life-table parameters derived from data collected from the colonized wild strains represent a maximum expression of their life-table parameters and are likely to reflect true differences between geographically isolated strains. Similar assumptions have been made to compare the life-table strategies of geographically distinct strains of *Culex quinquefasciatus*[13] as well as for carrying-on comparisons among populations of two triatomine species on three different studies[5–7] after colonization in the standard laboratory conditions.

Fig. 1: Locations where the populations were initially collected.

The studied colonies were maintained under conditions similar to those in a previously published study on the biology of *M. pallidipennis*[5]: at 27 ± 1°C and 75 ± 5% relative humidity. They were fed on immobilized and anesthetized New Zealand rabbits on a weekly basis. The rabbits were anesthetized following the Norma Oficial Mexicana regulations by administering 0.25 ml/kg of ketamine through intramuscular inoculation[14].

Methods

The eggs from each colony were grouped by the date of oviposition to initiate a cohort by population of 100 eggs each. After eclosion, the groups of each species of first-instar nymphs were separated individually into plastic containers (5.5 cm diam × 10.5 cm height) with a central vertical support of absorbent cardboard. Three days after eclosion, each cohort of nymphs was individually fed (as previously described) during a 1 h period; the subsequent blood meals were given weekly. The nymphs were observed at the end of feeding for recording of blood ingestion. The insects were maintained as previously described in a dark incubator with a 12:12 h light/dark photoperiod and were checked daily for ecdysis or death. Mortality was expressed as number of dead individuals divided by the initial number of individuals in the cohort.

At the end of the cycle, the percentages of female subjects in each studied cohort were recorded. From among the insects that completed development into adults, 10 adult pairs from each cohort were placed in individual containers (5.5 cm diam × 10.5 cm height) and maintained as previously described to determine the ovi-position patterns. Eggs were collected and counted daily for 90 days and placed in individual containers until hatching.

Statistical analysis

Descriptive analysis was performed among all variables. Percent mortality was calculated based on insect deaths per instar/initial number of individuals in the cohort. A non-parametric Kruskal-Wallis test was used to compare the amount of eggs laid per female and the developmental cycle periods in the three cohorts studied because the Bartlett's tests were *p* <0 for all comparisons. Pair wise comparisons were performed for intergroup comparisons using Dunn's method. A chi-square test was used to compare frequencies. The Sigma Stat 3.1 software (Version 3.1 for windows; Systat Software Inc., San Jose, CA) was used for statistical analysis. Whatever the probability level, it was restricted to a maximum of 5%.

RESULTS

The average egg-to-adult development time was significantly higher in the cohort from Luvianos (169.7 ± 19.6 days) than the average times from the other two studied populations, with no difference between these two (Table 1). A significant lower mean number of blood meals to molt (8.5 ± 1.44) was recorded for the cohort from Izúcar de Matamoros in respect of the two other studied cohorts (Table 1). Significant higher mean numbers of eggs were laid by female subjects daily from the cohorts from Chilpancingo de los Bravo (2.91 ± 1.41) and Izúcar de Matamoros (2.87 ± 1.1) than the cohort from Luvianos (Table 1). No significant differences were recorded when mortalities (from 23.6% in Chilpancingo de los Bravo to 29.7% in Luvianos), the egg eclosion rates (from 82.66% in Luvianos to 91.94% in Izúcar de Matamoros) or the percentages of female subjects at the end of the life cycles (from 53.85% in Izúcar de Matamoros to 57.81% in Luvianos) of the three studied cohorts were compared (Table 1).

DISCUSSION

The average egg-to-adult development time was shorter than five months in the studied cohorts from Chilpancingo de los Bravo and Izúcar de Matamoros. Those times were slightly longer than those for *Rhodnius colombiensis* and *Rh. prolixus* (144.4 and 117.7 days, respectively)[15] and those for *Triatoma rubida* (125.9 ± 3.3 days) from Chihuahua, Mexico[16]. In contrast, the average egg-to-adult development time was similar to those for the three populations of *M. pallidipennis* from central, southern and western Mexican states[5]. Those short average egg-to-adult development times mean a potential abundance of each studied population of *M. pallidipennis* on their respective distribution area. Those data match with the recorded high abundances on field collections of this species in many places (including Izúcar de Matamoros and Chilpancingo de los Bravo) in the states of Puebla and Guerrero[17–18], which could mean an increase in the risk of transmission of *T. cruzi* to the inhabitants of those areas.

On average, approximately 65–80% of instars in each of the study cohort from Luvianos and Chilpancingo de los Bravo required two or more meals to molt to the next instar, whereas the cohort from Izúcar de Matamoros required only one and a half meals to molt. The requirement for more blood meals represents an increased incidence of contact between the vector and hosts, which could lead to an increased infection rate by *T. cruzi* in humans, because *M. pallidipennis* usually defecates soon after feeding[19]. In contrast, the cohort from Izúcar de Matamoros required a less number of blood meals to molt, which could represent an advantage to survive in those environments where blood meal sources are scarce. A similar behaviour has been recorded for *T. protracta*, a desert triatomine[16].

The number of eggs laid per female per day (close to three eggs) after 90 days was significantly higher in the cohorts from Chilpancingo de los Bravo and Izúcar de Matamoros. Similar results were recorded for a cohort from a population of *M. pallidipennis* from the state of

Table 1. Biological attributes of *Meccus pallidipennis* from three different environmental areas

Characteristics	Mean ± SD			
	Luvianos (n = 91)	Chilpancingo (n = 89)	Izúcar de Matamoros (n = 90)	H $_{(d.f. = 2)}$
No. of development cycle (days)	169.7 ± 19.6[a]	144.4 ± 14.5[b]	141.7 ± 13.8[b]	51.49
Blood meals	12.4 ± 2.1[a]	11.7 ± 1.71[a]	8.5 ± 1.44[b]	44.95
No. of eggs laid per female per day	1.77 ± 1.1[a]	2.91 ± 1.41[b]	2.87 ± 1.1[b]	31.32
				χ^2 (d.f. = 2)
Mortality (%)	29.7[a]	23.6[a]	27.7[a]	0.97
Egg eclosion rate (%)	82.66[a]	90.7[a]	91.94[a]	2.45
Females (%) at the end of the cycle	57.81[a] (64)	54.41[a] (68)	53.85[a] (65)	0.37

Horizontally, numbers with similar superscript letters are not significantly different. Figures in parentheses are numbers.

Michoacán, western Mexico, reared under similar laboratory conditions to those on this current study[5]. The similarity of these three cohorts in that parameter reflects the high grade of adaptation of *M. pallidipennis* female subjects from the original populations from very different environmental areas to the study's controlled conditions. The egg eclosion rate was over 80% for the three studied populations (even over 90% in two cohorts) similar to *M. pallidipennis* in a previous study[5]. The mortality rates were low and not significantly different among the three studied cohorts and similar to those for *M. pallidipennis* in a previous study and *M. longipennis*, a related species[5–7]. The number of eggs laid per female per day, the high egg eclosion rate and the low mortality rates could mean an increase in the size of populations, leading to a potential increase in the risk of transmission of *T. cruzi* to hosts if those specimens get infected[5]. Results on these three parameters contributes to explain the high abundance of *M. pallidipennis* in the states of Michoacán, Puebla and Guerrero[2, 17–18] from western, central and southern Mexico, respectively. As reported for *M. pallidipennis* and *M. longipennis*[5–7], mortality in the youngest nymphs of *M. longipennis* appeared to be caused by the inability of the insects to feed because dead triatomines were generally found without significant intestinal content. In contrast, the mortality of older nymphs appeared to occur during molting.

At the end of the cycles, the proportion of female/male was close to 1:1 in the three studied populations. No significant differences were recorded when percentages of obtained female subjects were compared among studied populations. The abundance of female subjects leads us to conclude that apparently the three studied populations have the same potential to increase their abundance under favourable conditions, thus resulting in an increase in the risk of transmission of *T. cruzi* to hosts in their distribution areas. Similar results were obtained when three populations of *M. pallidipennis* were analyzed in a previous study as well as when four populations of *M. longipennis* were studied in two other researches[5–7].

Results from this current study were apparently influenced by the remarkable differences on the environmental conditions in the localities where the founders were initially collected. They also fit with those of a recent study where important differences on studied biological parameters among populations of *M. pallidipennis*[5] were recorded. These results on biological parameters are in coincidence with those for molecular and antennal phenotype analysis of populations of *M. pallidipennis* from different areas of Mexico[8].

CONCLUSION

The present study demonstrates the heterogeneity of the populations of *M. pallidipennis*, as well as that environmental characteristics from localities from where studied populations were initially collected affect on the average development time, on the mean number of blood meals to molt and on the mean numbers of eggs were laid by female. Based on the recorded data, the population from the hotter and wetter area (Izúcar de Matamoros) would represent a higher risk of transmitting *T. cruzi* to hosts. This study remarks the necessity of studies of local populations of each triatomine species, to know them better and to control them.

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