

Kissing Bugs. The Vectors of Chagas

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Abstract

A complete picture of Chagas disease requires an appreciation of the many species of kissing bugs and their role in transmitting this disease to humans and other mammals. This chapter provides an overview of the taxonomy of the major species of kissing bugs and their evolution. Knowledge of systematics and biological kinship of these insects may contribute to novel and useful measures to control the bugs. The biology of kissing bugs, their life cycle, method of feeding and other behaviours contributing to the transmission of *Trypanosoma cruzi* are explained. We close with a discussion of vector control measures and the allergic complications of kissing bug bites, a feature of particular importance in the United States.

8.1. INTRODUCTION

Vectors of Chagas disease are remarkable in being the only true bugs capable of transmitting disease to people. These bugs in the order Hemiptera differ from most other blood-feeding vectors by developing from egg to adult via nymphs that appear similar to adults but lack a pupal stage. All stages live in the same locations and exhibit nearly identical lifestyles. Most other insect vectors (lice are an exception) exhibit complete development including a pupal stage, thereby having dramatically different life stages and ecologies that can be targeted for control. All vectors of *Trypanosoma cruzi* are in the subfamily Triatominae, in the family Reduviidae, known as assassin bugs, so named because the majority of insects in this group are predators that hunt other arthropods and feed by digesting and sucking out their prey's bodily juices. These predators were presumed to have arisen from earlier plant feeding "sap suckers" (Carcavallo et al., 2000). The Triatominae are known as kissing bugs. Except for a group transported to seaports worldwide, Triatominae are restricted geographically to the American continents and probably emerged as a separate lineage coincident with the formation of the American continents.

Blood feeding evolved less than a dozen times in insects. Three groups of insects with a nymphal stage (bed bugs, lice and Triatominae) and another three groups with a pupal stage (fleas, flies and a butterfly) became hematophagous (Benoit and Denlinger, 2010). Most groups of blood-feeding insects are monophyletic (Yoshizawa and Johnson, 2010). However, in Diptera (flies), blood feeding is thought to have evolved at

least five times (Yuval, 2006). Triatomines are unusual arthropod vectors because they may harbour two trypanosome parasites with different modes of transmission, that is, faecal contamination for *T. cruzi* and salivary transmission for *Trypanosoma rangeli* (Gaunt and Miles, 2000).

The Triatominae contain an estimated 140 species; however, biological species are not always precisely defined (Knowles and Carstens, 2007) and some of the recognized groups likely contain multiple lineages, subspecies or morphological variants (Dorn et al., 2009). The family is subdivided into five tribes: Triatomini with nine genera and ~110 species; Rhodniini with ~20 species in two genera; Alberproseniini (one genus with two species), Bolboderini (four genera with 14 species) and Carvernicolini (two species in a single genus).

Although most Chagas transmission is vector-mediated via parasite-contaminated faeces, other routes include blood transfusion and organ transplant, and vertical transmission (i.e. mother to child) such that 2–10% of infants born to infected mothers are infected. Because the parasite cannot penetrate the skin, horizontal transmission from insect vector to mammalian host requires penetration of mucosal membranes (around the eye or mouth) or through the bite wound by parasites contained in faeces deposited while the vector is taking a blood meal. A second mode of horizontal transmission, thought to be rare in humans but perhaps common for other mammals, occurs when parasites penetrate the mucosa of the digestive system following ingestion of an infected vector (Diaz-Suarez, 2009).

8.2. VECTOR GENERA AND SPECIES

The relative importance of the various triatomine species as human disease vectors depends on behavioural and biological factors, especially the preferred vector ecotope with adaptation to domestic and peridomestic structures (corrals, chicken coops, wood piles, etc.) as well as the sylvatic habitat. Other important traits include degree of anthropophily and host preference, feeding time, dispersal capabilities and time span between blood meal and defecation (Lent and Wygodzinsky, 1979). The epidemiologically important vectors vary among geographic regions. Only about a dozen species appear to have strictly domestic populations over at least a part of their geographic range, while another two dozen or so species are described as secondary vectors because they seasonally or opportunistically invade houses from peridomestic and sylvatic habitats (Vallejo et al., 2009). The two most epidemiologically significant tribes include the Rhodniini and the Triatomini. Within the Triatomini, many taxa in the genus *Triatoma* have been organized into species groups called complexes.

8.2.1. North American taxa

Of the ~40 species found in North America, 8 are shared between Mexico and the United States, 4 are found only in the U.S. and 28 only in Mexico (some of these are also present in Central America) (Ibarra-Cerdena et al., 2009). Temperature is thought to limit the northern range to the lower approximately two-thirds of the United States. In Mexico, the species included in the phyllosoma complex are responsible for the majority of Chagas transmission (Ibarra-Cerdena et al., 2009; Martinez-Ibarra et al., 2008). These species are found associated with human dwellings in central and southern Mexico and include *Triatoma pallidipennis*, *T. longipennis*, *T. barberi*, *T. mazzotti*, *T. phyllosoma*, *T. picturata* and *T. mexicana*.

In the United States, transmission from insect vector to human host is rare and the major concern is anaphylactic reactions to their bites (Klotz et al., 2010). In Texas, *T. gerstaeckeri*, the principle vector of zoonotic *T. cruzi*, is associated with the southern plains woodrat, *Neotoma micropus* (Kjos et al., 2009). Other *Triatoma* species in Texas include *T. protracta*, *T. indictiva*, *T. rubida* and *T. neotomae*. In Arizona and California, *T. protracta* and *T. rubida* are commonly found, whereas *T. recurva* is rare. In the southeastern United States, *T. sanguisuga* has been found in large numbers around human dwellings and is associated with raccoons and opossums from Florida to Texas (de la Rua et al., 2010; Ponce, 2007).

8.2.2. Central American taxa

In Mesoamerica (roughly, central Mexico to Costa Rica), anti-Chagas campaigns have dramatically reduced the numbers of the introduced, domestic vector *R. prolixus* in most localities leaving the indigenous and widespread *Triatoma dimidiata* as the predominant vector except in Panama, where *R. pallescens* predominates. Additional minor species that may cause infection in humans include *T. nitida* and *T. ryckmani* (Ponce, 2007; Zeledón et al., 2007).

8.2.3. South American taxa

Chagas disease vectors in South America can be divided into three ecological groups, Andean, Amazonian and Southern Cone. The most significant domestic vectors in the Andes region, which includes part of five countries, Bolivia, Colombia, Ecuador, Peru and Venezuela, are *R. prolixus*, *T. dimidiata* and *Triatoma infestans*. Secondary vectors in this region are *T. maculata*, *R. ecuadoriensis*, *R. pallescens*, *T. venosa*, *T. carrioni*, *P. herreri*, *P. chinai*, *P. geniculatus* and *P. rufotuberculatus* (Guhl, 2007). It is in this region that *T. infestans* is thought to have originated in the Andean highlands and desert regions of Bolivia and Argentina (Giordano et al., 2005).

Deforestation and human settlement in the Amazon region have increased contact between humans and vectors. No species is reported to live year round strictly in human dwellings; however, several species are routinely associated with human dwellings including *T. maculata*, *P. geniculatus*, *R. neglectus* and *R. stali*. Humans also routinely have contact with several sylvatic species including *R. robustus*, *R. pictipes* and *R. brethesi* (Aguilar et al., 2007).

The Southern Cone region includes parts of Argentina, Bolivia, Brazil, Chile, Paraguay and Uruguay, and the major vector is *T. infestans*. Occasionally, *Panstrongylus megistus*, *T. brasiliensis*, *T. sordida*, *T. pseudomaculata* and *R. nasutus* are found (Dias, 2007).

8.2.4. The important human disease vectors

The most important human disease vector is *T. infestans*, followed by *R. prolixus*, and *T. dimidiata*. Different vector genera appear to be adapted to different niches with the result that the major subgroups of *T. cruzi* are associated with particular vectors and mammalian hosts (Gaunt and Miles, 2000). The major human disease vectors represent recent expansion into domestic and peridomestic habitats by only a few species. The habitat preferences of *Rhodnius* and *Triatoma* are reflected in their distribution within infested houses: *R. prolixus* is found in palm roofs and *T. infestans* in mud-walled houses or tiled roofs (Gaunt and Miles, 2000; Pizarro et al., 2008).

T. infestans occurs over a wide geographic range. Human dwellings and peridomestic structures are its preferred ecotope. Based on Zeledón's "defecation index", *T. infestans* adults are not quite as efficient as *R. prolixus* at defecating during feeding; however, both defecate within 10 min of feeding and this is thought to be the critical threshold for efficient *T. cruzi* transmission (Zeledón et al., 1977).

In the Andean highlands, *T. infestans* is found in rocky habitats in association with rodent burrows including wild guinea pigs (Pizarro et al., 2008). *T. cruzi* II appears to have evolved with *T. infestans* in rocky habitats or burrows associated with edentates or possibly ground dwelling marsupials (Gaunt and Miles, 2000).

R. prolixus is exclusively domestic in Central America. In northern South America, it occurs in domestic and sylvatic habitats, primarily palm trees (Lent and Wygodzinsky, 1979) where it feeds on opossums as well as birds. These bugs are well adapted to climbing on the smooth stems and trunks of palms. *T. cruzi* I predominates in domestic and sylvatic habitats in the regions north of the Amazon (Gaunt and Miles, 2000), and the sylvatic vectors are associated with opossums (Carrasco et al., 1996; Pova et al., 1984).

Studies examining local adaptation between vector and parasite strains are relatively few, but experimental studies have shown that not all *T. cruzi* strains can be transmitted by all species of Triatominae (Garcia and Azambuja, 1991). The insect vectors have a complex association with symbiotic intestinal bacteria, which may affect the numbers of the trypanosomatid parasites including *T. cruzi*, *T. rangelli* and *Blastocrithidia triatomae* (Azambuja et al., 2005; Eichler and Schaub, 2002).

8.3. EVOLUTION OF THE TRIATOMINES

The main evolutionary questions regarding Triatominae are as follows: Is this a monophyletic group, or alternatively, is it a polyphyletic group, having arisen from more than one predator ancestor? Did the Triatominae arise only in the New World or in both the New and Old Worlds? And finally, which species definition reveals the true taxonomic subdivisions? Answers to these questions give scientists the ability to infer shared characteristics, some of which, such as preference for human habitations and blood meals, are epidemiologically important.

8.3.1. Monophyly of Triatominae

Lent and Wygodzinsky (1979) concluded that the Triatominae subfamily are a monophyletic group possessing the following shared, derived characters: they are hematophagous, with a generally painless bite and have a membranous connection between the second and third rostral segments that allows them to bend the rostrum up during feeding.

The monophyly theory has been challenged based on several lines of evidence including biogeography and morphology, salivary proteins, cuticular hydrocarbons and DNA sequence differences between the Triatomini and the Rhodniini tribes (Dujardin and Schofield, 2004; Schofield and Galvao, 2009). When an insufficient number of shared characters or outgroups are used, the classification of monophyly or polyphyly suffers (Gorla et al., 1997; Schofield, 1988). So morphology, especially with a limited set of characters, may overestimate the differences among groups.

The differences in salivary proteins between Triatomini and Rhodniini are often cited as evidence for polyphyly (Ribeiro et al., 1998); however, other studies show that the anticoagulant mechanism utilized by the genus *Panstronglyus*, which is clearly within the Triatomini tribe (de Paula et al., 2005), is similar to that of *Rhodnius* and different from *Triatoma* (Pereira et al., 1996). Further, the differences in epicuticular hydrocarbons represent relatively primitive characters that differ quantitatively and not qualitatively (Juarez et al., 2002).

Results from many studies utilizing allozyme analyses and morphometry (Dujardin et al., 1999) and DNA sequence (Marcilla et al., 2001; Monteiro et al., 2000; Stothard et al., 1998) clearly show the separation of Triatomini from Rhodniini (Dujardin and Schofield, 2004). These studies used a single outgroup and showed the sister taxa relationship between the two tribes; however, with an insufficient number of outgroups, they cannot be used to prove or disprove mono- or polyphyly of the subfamily (Schaefer, 2003).

Cladistic analysis, with a large number (162) of morphological characters (Weirauch, 2008) or DNA sequences and a large number of outgroups (Weirauch and Munro, 2009), reveals the monophyly of the Triatominae subfamily. In addition, the power of this approach is that out of the 162 characters examined, Weirauch (2008) identified 12 shared characteristics that define the Triatominae subfamily. The best approach seems to be an evolutionary species concept using cladistic analysis with many characters, molecular as well as phenotypic and a sufficient number of outgroups to elucidate the phylogenetic relationships among taxa.

The evolution of the Triatominae from the Reduviidae ancestor appears to be fairly recent and originated in the Americas. This is supported by the lack of autochthonous species in Africa, the fact that the vast majority of species occur in the Western hemisphere (Dujardin and Schofield, 2004) and those found in Asia likely originated in the Americas (Hypsa et al., 2002). Dujardin and Schofield (2004) speculate they evolved “well after the mid-Tertiary period”, perhaps even later than the Quaternary period.

8.3.2. Evolution within the subfamily Triatominae

Among the epidemiologically important taxa are the previously mentioned sister taxa: Triatomini and Rhodniini. This division is well documented by phenotypic, morphometric and molecular (allozymes, DNA sequence, etc.) comparisons. The number of autosomes or sex chromosomes is not helpful in distinguishing the tribes, as the autosomal complement is nearly uniform (only three species have other than 20 autosomes). Genome size varies, and in general, Rhodniini has a smaller genome size correlating with smaller chromosomes rather than differences in the amount of heterochromatin (Panzeria et al., 2010). The other three tribes of Alberproseniini, Bolboderini and Carvernicolini have never been included in a formal cladistic analysis (Weirauch and Munro, 2009), and molecular genetic analysis to date has been limited to DNA regions coding for mitochondrial and nuclear ribosomal RNA (Patterson and Gaunt, 2010; Weirauch and Munro, 2009) and mitochondrial genes involved in electron transport (Patterson and Gaunt, 2010).

Within the Triatomini tribe, the *Triatoma* genus comprises ~80 species (Schofield and Galvao, 2009) that further subdivide into two clades found north and south of the Amazon region based on mtDNA (Lyman et al., 1999) and nuclear DNA sequence (Marcilla et al., 2001). More recently, a third clade has been added consisting of the dispar complex, located west of the Amazon region (Bargues et al., 2010). Cytogenetics is useful in distinguishing the two clades as all *Triatoma* in the “northern” clade, with one exception, show multiple sex chromosomes (fragmented X chromosomes), whereas most, but not all, “southern” clade *Triatoma* have the XY system (Panzer et al., 2010). The *Rhodnius* genus includes 16 species (Schofield and Galvao, 2009) that are divided into three clades based on mtDNA sequence (Lyman et al., 1999), allozyme analysis and morphometry (Dujardin et al., 1999), and they also are geographically localized.

Most workers have concentrated on understanding the evolution of *T. infestans*. The finding of wild foci of *T. infestans* in the Andean valleys of Cochabamba, Bolivia (Torrico, 1946) led to the hypothesis that this was the site of origin of the species (Schofield, 1988) and the vectors became domesticated as they moved from wild guinea pig burrows to quite similar microhabitats in human dwellings (Usinger et al., 1966). As might be expected from remnants of a founding population, results from early studies showed that although overall genetic diversity in *T. infestans* is low, these Andean *T. infestans* populations from Bolivia were more diverse than those found in Peru (Dujardin and Tibayrenc, 1985). Interestingly, some Argentinean populations show higher genetic diversity than is found in Bolivia (De Rosas et al., 2007; Garcia et al., 1995), presumably for reasons other than originating in Argentina, possibly from pesticide pressures (De Rosas et al., 2007). Chromosomal make-up is informative about the origin and dispersal of *T. infestans* as “Andean”. The putative ancestral form shows ~30% more DNA due to ~50% more heterochromatin than the “non-Andean” (Panzer et al., 2004). *T. infestans* appears to have lost heterochromatin as it spreads out across South America. Sequence of the internal transcribed spacer regions of the rDNA supports a northern/southern split in *T. infestans* populations (Bargues et al., 2006), and intermediate chromosomal forms have been detected in the overlapping area (Noireau, 2009; Panzer et al., 2007).

The interfertility of many species of the Phyllosoma (Mazzotti and Osorio, 1942) and Infestans complexes (Usinger et al., 1966) violates the traditional species definition of reproductive isolation (Mayr, 1970). Therefore, an ecological species concept (van Valen, 1976) of spatial or temporal division has been invoked for the separation of *T. infestans* and *T. platensis* that have some natural hybrids (Abalos, 1948) even though they occupy different niches, *T. infestans* in the domestic environment and *T. plantensis* in bird nests (Carcavallo et al., 2000). With the increasing

availability of DNA sequence data, quantitative differences in DNA sequence (genetic distance) measures may be useful in constructing taxonomic subdivisions (Monteiro et al., 2004; Pfeiler et al., 2006).

8.4. BIOLOGY OF THE BUGS

The Triatominae are a nearly obligatory hematophagous subfamily, although some species feed on the hemolymph of arthropods in addition to vertebrate blood and appear to represent the ancestral trait of the subfamily (Sandoval et al., 2010). They are relatively large bugs with adults ranging in length from about 12 to 36 mm and having five immature instars, morphologically similar to adults, except lacking wings. Both sexes and all five instars feed on blood of vertebrates, starting with the first instars which take a blood meal as soon as 2–3 days after hatching from the egg. All instars are also capable of transmitting *T. cruzi*, though first instars probably rarely transmit the parasite as they must acquire the parasites from an initial blood meal and often moult to second instars before feeding again.

The life cycle of kissing bugs can be completed in several months in tropical areas where temperatures are warm and when food is readily available. In temperate higher latitude regions having extended cooler periods of the year, the bugs typically have only one generation per year. In these environments, bugs seek refuge in cavities, rodent burrows, caves or similar locations with moderate temperatures and pass the unfavourable times in various nymphal stages. Adults live several months, occasionally a year or more (Gorla et al., 2010) and feed throughout their lives. In captivity, adults of *T. rubida*, *T. protracta*, and *T. recurva* live 4–5 months on average (J. Schmidt, unpublished data). Mating is of short duration relative to many other hemipterans, lasting only about 10 min. Females readily mate with multiple males and can lay several hundred eggs during their lifetime, but numbers vary widely depending on conditions including density of bugs in the immediate environment, presence of preferred hosts, continuous availability of hosts and time of season the bug matured (Gorla et al., 2010). Dispersal is achieved both actively by crawling immatures and flying adults (Schofield, 2000) and passively in some species either by transport of attached eggs to new locations or by human transport of materials harbouring eggs or free-living stages. A classic example of human passive transport is the species *T. rubrofasciata* which has been inadvertently transported via commerce to many seaports around the world (Schofield, 2000). Active dispersal of immatures appears to be in response to a shortage of food sources leading to bugs crawling towards locations potentially having new hosts. Adults typically disperse by flight during optimal seasons, often just before or

during the rainy season. Hunger is a powerful driver of dispersal (Lehane et al., 1992). Another is the reproductive strategy in which gravid females retain a portion of their eggs for deposition after flight (Klotz et al., 2010; McEwen and Lehane, 1994). Mate seeking is apparently not a cause of dispersal as evidenced by nearly equal numbers of males and females being captured at lights and the captured females rapidly ovipositing fertile eggs (Klotz et al., 2010; McEwen and Lehane, 1994).

Triatomine bugs can inhabit a variety of ecological environments from highly specialized to generalized, including human domestic environments. Within these habitats, they can range from specialists feeding on a limited number of hosts or to opportunists feeding on a variety of host species. Specialists include *Cavernicola pilosa*, which feed on bats, the members of *Psammolestes* living in twig constructed nests of birds, and *Rhodnius* spp. living in palm trees where they feed on birds and mammals. Living in somewhat more general areas are *T. rubrofasciata* and members of the *T. protracta* complex that prefer feeding on rodents. *T. infestans*, *T. dimidiata* and *R. prolixus* are inhabitants of human domestic environments (Lehane et al., 1992). Most species, irrespective of their preferred habitat, can be opportunistic and feed on available species when encountered. Species that are generally opportunists can also be facultative specialists that prefer to feed mainly on one group of hosts if given the opportunity (de la Fuente et al., 2008; Gurtler et al., 2009; Pizarro and Stevens, 2008). The *Triatoma* living in Arizona are all non-domestic species that often feed on pack rats (*Neotoma* spp.), yet they readily feed on a variety of encountered vertebrate hosts including humans (L. Stevens et al., unpublished data). In Tucson, the Arizona-Sonora Desert Museum, an educational living zoo of regional animals, supports large populations of *T. rubida* and *T. protracta* which plague many of the captive animals. Their extensive feeding is considered the cause of anaemia in several reptiles and the death of a rare protected mountain rattlesnake (James L. Jarchow, DVM, personal communication).

Exactly how and when some species of triatomines became specialists on humans is unclear. Human beings are relatively recent inhabitants of the New World, with exact arrival dates debated, but arrival around 14,000–20,000 years ago is reasonable. Thus, the shifts in some triatomine species to specializing on our species are evolutionarily recent events and might well explain some of our vulnerability to the *T. cruzi* parasites the bugs transmit to us.

8.4.1. Feeding behaviour

Triatomine bugs rely on a multitude of sensory modalities to locate hosts. These include carbon dioxide (CO₂) gradients, odours, moisture, heat and air flow (Guerenstein and Lazzari, 2009). They are exquisitely sensitive to

CO₂, being able to detect 75 ppm above the background air levels of 350 ppm when a trace of lactic acid, another attractant, is present. Exothermic hosts such as resting reptiles generate very low levels of CO₂; nevertheless, triatomines use many other host-generated chemical cues including short-chained aliphatic amines and acids, 7- to 9-carbon aldehydes and alcohols and lactic acid to locate these hosts. Triatomines also detect very low infrared radiation enabling them to orient towards endothermic hosts from several metres. Finally, they follow air currents impregnated with host odours to the host (Guerenstein and Lazzari, 2009).

Feeding by bugs in human domiciles usually takes place at night, with bugs secreting themselves during the day in cracks and crevices away from the feeding areas. Much less is known about feeding times for sylvatic species. Likely, they adjust their daily activity periods to match the inactive periods of their host, thereby minimizing danger and interference from hosts. *T. rubida* and *T. protracta* do not hesitate to approach and feed on rodents during bright daylight (Klotz et al., 2010), a behaviour that might well be adaptive for bugs that often feed on rodents in their burrows while they are sleeping or quiescent during the day.

Once a host is located, a hungry bug extends its proboscis and inserts it into the skin of the host. Often a host detects the minor sensation caused by a bug's probing and moves or shifts position, causing the bug to retract before returning to continue probing one or several more times until feeding is complete. Uninterrupted feeding is relatively rapid—considering that the bug often imbibes more blood than its own weight (Klotz et al., 2010) and feeds on average 22 min for *T. protracta*, 28 min for *T. rubida* and between 11 and 28 min for *T. infestans*, *T. dimidiata* and *R. prolixus* (Klotz et al., 2009b; Zeledón et al., 1977). To rid their bodies of the excess fluids from their blood meals, bugs often defecate during feeding, at completion of feeding, or after leaving the feeding site. Some species, especially those domestic species that are major vectors of Chagas disease, defecate rapidly, frequently, often near or on the host (Zeledón et al., 1977). Others not considered important vectors tend to defecate less rapidly and frequently, and often at a distance from the host (Klotz et al., 2009b). Frequency and proximity of defecation to the host are considered factors in transmitting *T. cruzi* to humans.

8.5. URBANIZATION OF THE BUGS

Many species of bugs like *T. dimidiata* occupy multiple ecotopes including sylvan, peridomestic and domestic environments providing reservoirs to readily occupy treated houses. For example, *T. mexicana* of central Mexico and *T. dimidiata* of the Yucatan, Mexico and Belize do not colonize homes

but seasonally migrate from sylvan and peridomestic ecotopes and infest homes (Ramirez-Sierra et al., 2009). Thus, the efficacy of insecticide programmes is greatly reduced as new populations of triatomines infest homes annually. Molecular markers have been used to determine the source of bugs in houses after treatment and whether they are survivors of ineffective pesticide applications (the usual case) or are immigrants from a particular ecotope. A number of studies of vector-borne infectious diseases demonstrate that their resurgence is due to human expansion into forested areas and Chagas disease in Latin America follows this paradigm. For example, the diversity of *T. dimidiata* niches defies public health initiatives because of continued reinfestation from sylvatic habitats posing a greater threat than *R. prolixus* even though *R. prolixus* is more efficient at transmitting disease (Ponce, 1999). Other examples include niche expansion into houses in a forested community in Bolivia and homes in Calilegua National Park, Argentina where light attracts *P. rufotuberculatus* (Salomon et al., 1999).

8.6. CONTROL OF VECTORS

8.6.1. Multinational efforts

Vector transmission of Chagas disease accounts for more than 80% of disease transmission (Yamagata and Nakagawa, 2006). Over the past two decades, several regional initiatives in Latin America have made tremendous strides in reducing and in some cases, interrupting vector transmission of the disease. The Southern Cone Initiative was launched first (1991) and focused on blood screening and insecticide-based elimination of *T. infestans*, a primarily domestic species that is the main disease vector in the southern cone region of South America. Success, although considerable, was measured mainly by the significant reduction of *T. infestans* in Uruguay by 1997, Chile by 1999, Brazil by 2000, Argentina by 2001 and Paraguay by 2002. Bolivia was the only country that did not see a significant reduction of the primary vector, *T. infestans*.

The Central American Initiative followed (1997) with the goal of eliminating *R. prolixus*, a South American species with only domestic populations in Central America and reducing household infestations of *T. dimidiata*, a native domestic, peridomestic and sylvatic species (Ponce, 2007). Other regional initiatives were launched in the Andean Region (1997), Mexico (2003) and Amazon (2004). To date, Brazil, Chile, Uruguay and Guatemala have been declared free of Chagas disease transmission by their main vectors, as well as some areas in Argentina and Paraguay (Gorla et al., 2010).

Large-scale initiatives like these face formidable challenges. The operational logistics of controlling vectors over vast geographic areas should be designed to maximize efficacy and minimize costs (Levy et al., 2010). The sustainability of these programmes relies on political and economic commitments that may change quickly in developing countries, and the decentralization of health care services across Latin America during the 1980s places the burden of responsibility on vector control and surveillance on the local community (Gürtler et al., 2007; Tarleton et al., 2007). Although there exists technical expertise in management strategies, there is always room for improvement and yet applied research in vector control lags far behind basic research on triatomines (Abad-Franch et al., 2010). Finally, a successful control campaign can often be its own worst enemy if complacency sets in and vector control and surveillance programmes are not maintained (Gorla et al., 2010).

8.6.2. Housing improvement

Vector control programmes for triatomines generally include three components that should be carried out by trained personnel (Schofield, 2000): (1) geographical reconnaissance to locate infestation foci, which entails inspections of homes and peridomestic environments in areas suspected of infestation; (2) application of residual insecticide sprays in and around infested structures and (3) long-term surveillance to monitor treated homes and peridomestic environments for reinfestation and retreatments when necessary. Homeowners should also be educated on the vector control programme and encouraged to report any triatomines that they find.

8.6.3. Chemical control

Insecticide spraying has been the cornerstone of vector control of triatomines ever, since the advent of organic insecticides in the late 1940s made effective control possible. The organochlorines such as dieldrin and lindane were the first line of defense in the 1950s, followed by organophosphates and carbamates in the 1960s and 1970s, and pyrethroids since the 1980s, such as cypermethrin, cyfluthrin, deltamethrin and lambda-cyhalothrin, particularly wettable powder and soluble concentrate formulations (Gorla et al., 2010; Toloza et al., 2008). Ideally, chemical control measures should be tailored to the biology of the targeted species, as for example, with *T. dimidiata* in the Yucatán Peninsula, by spraying at the beginning of April, right before the start of their seasonal infestation period (Barbu et al., 2009).

Chemical failures due to pyrethroid resistance have been reported in areas of Argentina and Bolivia in controlling *T. infestans* including evidence of resistance in eggs to the pyrethroids deltamethrin and lambda-cyhalothrin (Tolozza et al., 2008). Resistance may be slowed by rotating or mixing carbamates or organophosphates with pyrethroids (Lardeux et al., 2010). Other chemical control strategies include pyrethroid-impregnated curtains (Ferral et al., 2010) and bed nets (Kroegeer et al., 2003), and xenointoxication, as for example, using pyrethroid-treated dog collars (Reithinger et al., 2006). Because young children spend much of their time in and around their household, efforts need to consider the effect of insecticide sprayings in domestic settings (Chaudhuri, 2004).

8.6.4. Non-chemical measures

Chagas disease is considered one of the neglected tropical diseases that have primarily impacted the poorest people living in Latin America (Hotez et al., 2008). Early on, Carlos Chagas recognized the potential benefits of improved housing in the prevention of triatomine infestation (Dias, 2007). According to Zeledon and Rabinovich (1981), some of the risk factors associated with triatomine infestations include “palm thatched roofs or walls” for *R. prolixus*, “cracks in mud and cane walls” for *T. infestans*, and “wooden or mud houses with earthen floors or under houses built on stilts” for *T. dimidiata*. Replacing thatched roofs with metal sheets (Bustamante et al., 2007) and plastering walls (Monroy et al., 2009) removes potential harbourage sites for triatomines, as well as physical modifications in the peridomestic environment such as replacing piled brushwood with wire fencing for animal enclosures (Gürtler et al., 2007).

Investigations of potential biological control agents have been undertaken in order to provide alternative strategies to chemical control measures and/or management of pyrethroid-resistant populations of triatomines (Pedrini et al., 2009). The entomopathogenic fungus, *Beauveria bassiana*, has been isolated from field populations of *T. infestans* in Argentina. Pathogenicity tests of these isolates in the laboratory yielded 100% mortality of bugs 15 days after infection (Marti et al., 2005). Field tests of traps infected with *B. bassiana* were conducted in rural village homes infested with pyrethroid-resistant bugs, and their mortality was estimated at >50% (1). Other pathogens isolated from *T. infestans* include a picorna-like virus (Muscio et al., 1988). Ryckman and Blankenship compiled a list of a number of potential parasites and predators of the Triatominae. How practical and where in the life cycle of the kissing bugs these biological agents could be used remains to be established (Ryckman and Blankenship, 1984).

BOX Non-chemical measures to reduce risk of household infestation**Sanitation measures**

- Maintaining good hygiene and reducing clutter inside houses (Monroy et al., 2009)
- Eliminating junk, wood or rock piles, trash, and other objects in peridomestic environments (Ferral et al., 2010)
- Managing surrounding vegetation and conserving local fauna (Teixeira et al., 2009)
- Home improvement
- Installing insect screens (Guzman-Tapia et al., 2007)
- Replacing outside white lights with yellow lights (Gouge et al., 2010)
- Sealing structural cracks and crevices (Gouge et al., 2010)

8.7. NON-INFECTIOUS CONSEQUENCES OF A TRIATOMINE BITE

The major importance of kissing bugs from a medical perspective is their ability to transmit *T. cruzi* to humans and other animals, including pets. The transmission of *T. cruzi* remains an ongoing problem in Mexico, Central and South America. An unappreciated health-related association important in the United States is allergic responses to kissing bug bites. The allergic reactions can be trivial to life-threatening (Klotz et al., 2010). For example, anaphylaxis caused by a *Triatoma* bite led to the death of an adult woman in Phoenix, Arizona (Lo Vecchio and Tran, 2004). *Triatoma* bites are the leading cause of anaphylaxis due to insect bites in the United States (Hoffman, 1987).

Kissing bugs rarely transmit Chagas in the United States. Why this is so is unknown and is an active area of interest. To date, only six autochthonous cases of Chagas are documented in the United States (Dorn et al., 2007). Lack of domestication of sylvatic species is likely the most important factor (Zeledón, 1974). Observers have further postulated that because Triatominae in the United States do not defecate at the time of feeding there is little or no transmission of *T. cruzi* (Edwards and Lynch, 1984). The latter speculation is borne out by direct observations of *T. sanguisuga* (Shields and Walsh, 1956) feeding on humans and *T. rubida* and *T. protracta* feeding on rodents (Klotz et al., 2009b; Wood, 1951). It is interesting to note that victims of kissing bug bites in the United States do not report faecal drops or smears on their bed sheets (Shields and Walsh, 1956) yet defecation while feeding or shortly after the insect is replete is very characteristic of other Triatominae.

8.7.1. Nature of the bite

There is considerable misinformation concerning kissing bug bites, in particular, whether the bite is painful or not. Early accounts of bites of *T. sanguisuga* stated the bite is "...very painful and often even dangerous" (Kimball, 1893–1894) and "...a most painful wound" often confused with a spider bite (Le Conte, 1854–1855). From the standpoint of successful hematophagous insects, it is helpful that the host remains unaware of the presence of the insect; hence, the act of biting is often painless. Patients presenting to emergency rooms with anaphylaxis due to kissing bugs bites are rarely aware of a preceding bite—they awaken with intense itching over the body and difficulty breathing (Shields and Walsh, 1956). Shields and Walsh carefully observed *T. sanguisuga* feeding on humans and concluded that the bite is painless (Nichols and Green, 1963).

The typical kissing bug bite leads to a small raised skin lesion with a central punctum and an inflammatory infiltrate in the subcutaneous tissue (Shields and Walsh, 1956). Swelling of tissue at the bite site over several days may lead to nodular, urticarial lesions associated with pain and may last up to 7 days (Lynch and Pinna, 1978). Some severe reactions may last for up to a month and include lesions at the bite site. Lymphadenitis and vasculitic rashes such as erythema multiforme may occur as well (Shields and Walsh, 1956). These delayed allergic responses are uncomfortable or painful and undoubtedly account for the many claims that the bite itself "hurts". Romana's sign (unilateral eyelid oedema), found in patients in endemic areas of Chagas, is held to be pathognomonic of acute disease, but when seen in the United States is likely to be an allergic manifestation of the bite (Klotz et al., 2010).

Allergic reactions to bites of many species including the epidemiologically important vectors of Chagas disease (*T. infestans*, *R. prolixus*, *P. megistus* and *T. dimidiata*) are documented (Mott et al., 1980). The use of xenodiagnosis has been limited as a result of allergic skin reactions occurring after bites—patients experienced accelerated allergic reactions such as wheals and hives (i.e. IgE-mediated responses; Costa et al., 1981).

8.7.2. Triatomine saliva and its antigenic components

Whole bug extracts of *T. infestans* are capable of eliciting IgE and IgG antibodies in atopic patients, and the allergens may be similar to those of the American cockroach, *Periplaneta americana* (Alonso et al., 2004). However, mere contact of kissing bugs with human skin has not been reported to cause allergic reactions such as are seen with, for example, the urticating hairs of some caterpillars (Klotz et al., 2009a).

Allergic host responses to kissing bug bites are due to allergens present in the saliva of the bugs. In kissing bug endemic areas in the United

States, allergies are common. About 7% of the inhabitants of one community in California had evidence of IgE allergy to *T. protracta* (Marshall et al., 1986b). Bioassays of salivary gland extracts have demonstrated the presence of allergens capable of eliciting IgE antibody in patients and are highly species specific, that is, there is no cross-reactivity of extracts, for example, between *T. rubida* and *T. protracta* or extracts from *R. prolixus*, *T. rubrofasciata* or *T. cavernicola* (Marshall et al., 1986a). The allergens appear to be low molecular weight proteins (Marshall et al., 1986a). Procalin, a 20-kDa protein, was identified from *T. protracta* (Paddock et al., 2001). This protein, or one very similar, accounted for ~90% of the allergenic activity of the saliva (Chapman et al., 1986).

In the classical description of anaphylaxis by Richet and Portier, two steps were required: exposure to an allergen (which “primed” or “set the stage”) followed by repeat exposure to the same allergen *after* sufficient time (usually weeks) for lymphocytes to produce IgE antibody to the foreign antigen and thus elicit the allergic cascade known as a type I hypersensitivity reaction (Klotz et al., 2009a). This reaction is the most extreme of allergic reactions mediated by IgE antibody. During the reaction, antibodies bind to mast cells located throughout the body and cause the release of histamine which in turn leads to swelling of the mucosa especially in the airways and larynx that can impair breathing, syncope, hypotension and shock, urticaria (hives), intense itching, diarrhoea and even uterine bleeding (Nichols and Green, 1963; Pinnas et al., 1986). IgG antibodies may result in a similar, but not identical response known as an anaphylactoid reaction, but this has not been reported with kissing bug bites and usually follows some form of envenomation (Klotz et al., 2009a). Anaphylaxis has been reported following bites with important Chagas vectors including as *R. prolixus* and *T. infestans* but is apparently not common (Mott et al., 1980).

Currently, there is no effective preventive therapy for anaphylaxis due to kissing bug bites. Moffitt et al. (2003) compiled the data regarding immunotherapy and its use and success in patients with a history of anaphylaxis, but this is an intensive procedure and without standardized allergens it is unlikely to be of much widespread benefit.

8.8. CONCLUSION

Extensive research on the primary vectors of Chagas has provided scientists with a wealth of knowledge regarding the origin and expansion of domesticated triatomines and has led to many successful control programmes. However, secondary vectors that are often cryptic in nature and enjoy a relatively large niche in forests and burrows readily move into vacated ecotopes and pose an ongoing risk for transmission of *T. cruzi*.

Consequently, kissing bugs will remain a menace to human populations for some time to come as well as a subject of intense research.

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