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Research Article

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House Reinfestation With *Triatoma infestans* (Hemiptera: Reduviidae) After Community-Wide Spraying With Insecticides in the Argentine Chaco: A Multifactorial Process

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Abstract

We investigated the dynamics and underlying causes of house (re)infestation with *Triatoma infestans* (Klug 1834) after a community-wide residual spraying with pyrethroids in a well-defined rural section of Pampa del Indio municipality (northeastern Argentina) over a 4-yr period. House infestation was assessed by timed manual searches, during insecticide applications, and by opportunistic householders' bug collections. All reinfested houses were selectively re-sprayed with insecticides. The resident population comprised Qom (66.6%) and Creole (33.4%) households, whose sociodemographic profiles differed substantially. The prevalence of house infestation dropped, less than expected, from 20.5% at baseline to 5.0% at 14 months postspraying (MPS), and then fluctuated between 0.8 and 4.2% over 21–51 MPS. Postspraying house infestation was positively and highly significantly associated with prespraying infestation. Most of the foci detected over 14–21 MPS were considered persistent (residual), some of which were moderately resistant to pyrethroids and were suppressed with malathion. Infestation patterns over 27–51 MPS suggested bug invasion from internal or external foci, but the sources of most findings were unaccounted for. Local spatial analysis identified two hotspots of postspraying house infestation. Using multimodel inference with model averaging, we corroborated that baseline domestic infestation was closely related to refuge availability, housing quality, and occurrence of peridomestic infestation. The diminished effectiveness of single pyrethroid treatments, partly attributable to moderate resistance compounded with rather insensitive vector detection methods and poor housing conditions, contributed to vector persistence. Improved control strategies combined with broad social participation are needed for the sustainable elimination of vector-borne human Chagas disease from the Gran Chaco.

Key words: *Triatoma infestans*, Chagas disease, vector control, reinfestation, insecticide resistance

Neglected tropical diseases (NTDs) affect resource-constrained, vulnerable populations, which usually have scarce visibility and low priority (Hotez et al. 2008). Chagas disease is one of the NTDs that cause most serious health problems in Latin America, where it ranks among the most important infectious diseases in terms of disability-adjusted life-years (Lee et al. 2013). Argentina, Brazil, and Mexico are believed to have the highest number of infected people according to the World Health Organization (WHO) estimates based on 2010 data (WHO 2015). Vector-borne transmission of human infection with *Trypanosoma cruzi* (Chagas 1909) is still considerable in the Gran Chaco eco-region, including sections of Argentina, Bolivia, and Paraguay (WHO 2015). Moreover, international migration driven by socioeconomic and political factors has expanded Chagas disease to nonendemic areas and developing countries (Schmunis and Yadón 2010). The total annual cost of an infected person to society (including healthcare costs and productivity losses) may range from US$4,000 in Latin America to US$13,000–15,000 in Europe, North America, and Oceania (Lee et al. 2013). Therefore, prevention of vector-borne transmission combined with etiologic treatment of infected patients and newborns in traditionally endemic countries contribute toward reducing the global burden and societal cost of Chagas disease.

The main vector of *T. cruzi* in the Southern Cone countries and southern Peru is *Triatoma infestans* (Klug 1834), which traditionally thrives in domestic and peridomestic habitats of rural houses and (peri)urban settings (WHO 2002, Bayer et al. 2009, Provecho et al. 2014). Therefore, Chagas disease vector control has mainly been based on the residual application of insecticides to (peri)domestic habitats since the late 1940s. Sylvatic foci of *T. infestans*, well-known in Bolivia for decades (Noireau et al. 2005, Brenière et al. 2013), were later discovered in Chile, Argentina, and Paraguay (Ceballos et al. 2009, Bacigalupo et al. 2010, Rolón et al. 2011). Nonetheless, insecticide-based campaigns strongly reduced vector-borne transmission of human *T. cruzi* infection and reduced the geographic range of *T. infestans* throughout the Southern Cone (Dias et al. 2002, Schofield et al. 2006). However, indices of house infestation with *T. infestans* still remain high in the Gran Chaco region and domestic transmission persists, albeit at lower levels (Porcasi et al. 2006, Gürtler et al. 2007, Gurevitz et al. 2013, Cardinal et al. 2014, Gaspe et al. 2015a).

House infestation with triatomine bugs is determined by several processes that include social, ecologic, environmental, and cultural factors frequently operating in a framework of chronic rural poverty and weak healthcare services (Gürtler 2009, Dumonteil et al. 2013, Bustamante et al. 2014, Gürtler and Yadon 2015). This combination poses serious challenges to the effectiveness and sustainability of vector control programs. The situation in the Gran Chaco appears to be complex owing to the interplay of several factors: the much lower effectiveness of pyrethroid insecticides in peridomestic habitats (Gürtler et al. 2004, Cecere et al. 2013), the dispersal capacity of *T. infestans* (Vázquez-Prokopec et al. 2006, Abrahan et al. 2011), the eventual occurrence of sylvatic foci (Ceballos et al. 2009, Rolón et al. 2011), persisting rural–urban migration (Briceño-León and Méndez Galván 2007), poor housing quality and related sociodemographic features (Dias and Dias 1982, Cecere et al. 1998, Gurevitz et al. 2011, Gaspe et al. 2015a), and frequent reports of pyrethroid resistance in *T. infestans* (Mougabure-Cueto and Picollo 2015). Therefore, more evidence on how to improve the effectiveness of vector control efforts in the Gran Chaco region is greatly needed.

As part of a longitudinal program on the eco-epidemiology and control of Chagas disease in this region, the current study was conducted in Pampa del Indio municipality, a highly endemic area of the Argentine Chaco inhabited by creoles and an indigenous group (Qom or "Toba"). The same protocol of intervention was successively applied to three large rural sections of the municipality encompassing 300–400 houses each (Areas I, II, and III). Preintervention house infestation rates in Areas I and III ranged from 30 to 40% (Gurevitz et al. 2011, Gaspe et al. 2015a). Domestic infestation was mainly associated with the physical structure of the house (refuge availability), householders' use of domestic insecticides, residential overcrowding, distance to the nearest infested house, and household level of educational attainment. The impact of community-wide spraying with pyrethroids remarkably differed between Area I (where early vector control failures were attributed to moderate levels of pyrethroid resistance, Gurevitz et al. 2012) and the adjacent Area III, where postintervention house infestation was <1% over a 4-yr follow-up (Gaspe et al. 2015b). Here we focus on Area II, which differed in several aspects from our previous study locations, to describe the spatiotemporal dynamics of house (re)infestation with *T. infestans* over a 4-yr period after community-wide spraying with pyrethroids. Specifically, we reexamined the significance of the putative sources of house reinfestation identified earlier (i.e., residual foci that survived pyrethroid treatment and house invasion from external foci, such as nontreated peripheral areas) and whether pyrethroid resistance played any role in apparent vector control failures; identified processes and factors related to house infestation; and examined the predictive power of an earlier model of house infestation developed for Area III.

Materials and Methods

Study Area

This study was conducted in five rural communities (Lote Cuatro, Campo Nuevo, Campo Medina, Cancha Larga, and Pampa Ombú) spread over 300 km^2 in the southeastern section (hereafter denominated Area II) of Pampa del Indio municipality (25° 55′ S 56° 58′ W), Chaco province, Argentina (Fig.1). The environmental and socioeconomic characteristics of Pampa del Indio were described by Gaspe et al. (2015a). The study area had last been sprayed with insecticides by Chagas disease control programs in 1997–1998. In total, 34 houses from Lote Cuatro were sprayed with pyrethroids by local hospital staff in late 2006; hospital records of house infestation and insecticide treatment showed that 26 (77%) of them had been infested before insecticide spraying.

Study Design

All houses in the area were registered during July–September 2008 (late winter–early spring), and a systematic cross-sectional survey of house infestation was carried out in one every three houses in three communities (including 85.6% of all Area II houses). All houses were inspected in the remaining two communities (Cancha Larga and Pampa Ombú). Immediately after the baseline study was completed, a community-wide insecticide spraying campaign aiming at full coverage was undertaken. In total, 127 (34.0%) inhabited houses from Campo Medina, Lote Cuatro, and Campo Nuevo were inspected for the presence of triatomine bugs in July 2008, whereas 58 (92.1%) inhabited houses from Cancha Larga and Pampa Ombú were inspected in September 2008.

Monitoring of postspraying house infestation was conducted in October 2009 (14 months postspraying, MPS), May 2010 (21 MPS), December 2010 (27 MPS), December 2011 (39 MPS), and December 2012 (51 MPS). Houses infested with *T. infestans* were selectively sprayed with insecticide immediately after each survey. A sociodemographic and environmental household questionnaire was conducted at every postspraying survey (Supp. Table 1 [online only]).

Household Surveys

All house compounds and public buildings existing at baseline were identified with a numbered aluminum plate to facilitate subsequent re-identification, and their location was georeferenced with a GPS receiver (Trimble GEO XM, Trimble Inc., California, USA; eTrex Legend HCx, Garmin Internacional, Inc. Kansas, USA) after obtaining the express consent of each household head. A household is defined as the group of people residing in a house compound and includes all related and unrelated family members. A house compound encompasses one or more domestic premises (either separate or adjacent sleeping quarters) and several peridomestic structures.

The intervention objectives were explained to each household head on our first visit. Householders were asked for the presence of triatomine bugs on house premises at each survey. Dry specimens of *T. infestans*, *Triatoma sordida* (Stal 1859), and other Reduviidae were shown to them to avoid confusion with other insects. Householders were then provided with a labeled self-sealing plastic bag to place any triatomine they may sight and were instructed on how to safely collect and keep them. Householders were asked for any triatomine at each follow-up survey to derive two indices: householders' notification of house infestation and householders'

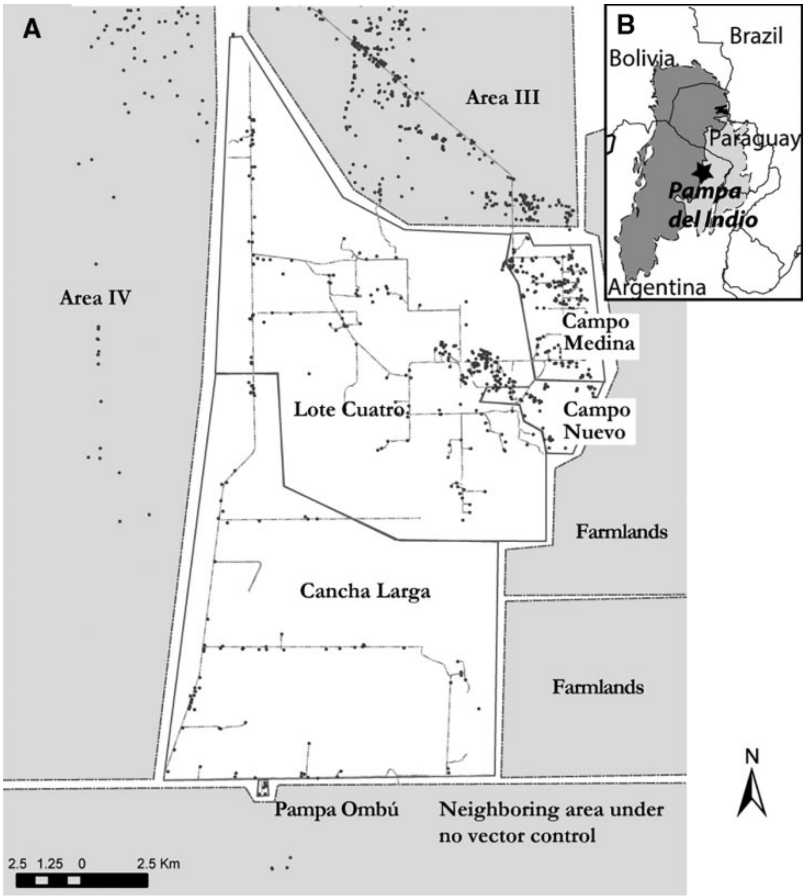

Fig. 1. A. Map of the study area (Area II, in white) and adjacent rural communities (in gray) within Pampa del Indio municipality, and neighboring districts under no regular vector control. B: Location of Pampa del Indio, Chaco, Argentina, showing the humid (darker gray) and dry (lighter gray) Chaco ecoregions.

bug collection, respectively. Householders' notification of the domestic presence of *T. infestans* and timed searches of domestic infestation were compared using the kappa index implemented in Stata 12 (Stata Corp 2012, College Station, TX). Kappa index values greater than 0.6 are taken to represent substantial to perfect agreement, whereas values less than 0.4 are taken as poor agreement.

All (peri)domestic sites in each house compound were registered and located on a sketch map at each survey. Information on environmental, demographic, and socioeconomic factors putatively associated with house (re)infestation with *T. infestans* was collected at 14, 21, 27, 39, and 51 MPS surveys. Most of the selected variables were based on background evidence indicating they were associated with house infestation (Gurevitz et al. 2011, Saunders et al. 2012, Gaspe et al. 2015a): ethnic background, number of resident people (household size), indoor presence of domestic animals, livestock number, source of light, and total number of bedrooms (used to calculate an overcrowding index: number of people per bedroom). We computed a goat-equivalent index to quantify the total number of livestock (cows, pigs, goats) and poultry owned by each household expressed in terms of goat biomass, assuming average weights for cows (453 kg), pigs (159 kg), goats (49 kg), and chickens (2.5 kg; Ministry of Agriculture, 2010). Housing quality (a three-level categorical variable) was represented by the combination of mud walls (vs. brick-cement walls) and tarred-cardboard sheets on the roof (vs. corrugated metal-sheets); no house had brick-cement walls and tarred-cardboard sheets. We also recorded the occurrence of wardrobe and boxes for clothing storage, reported use of domestic insecticides, type of construction materials used in domestic premises, and a refuge availability index for *T. infestans*, as described by Gurevitz et al. (2011).

Vector Surveys and Spraying Operations

Timed manual searches of triatomines were conducted in all (peri)domestic sites within a given house compound by two skilled bug collectors using a dislodging aerosol (0.2% tetramethrin; Espacial, Argentina), as described by Gurevitz et al. (2011). Each (peri)domestic site was inspected by one person during 15 min. Inhabited houses that were closed on our first attempt to inspect them were re-visited on —one or two occasions at times recommended by the nearest neighbors.

A house compound was considered infested if at least one live *T. infestans* (except eggs) was found by any collection method (i.e., timed manual searches, during insecticide spraying operations, and householders' bug collections) in any (peri)domestic site of the inspected houses. Domestic infestation refers to the finding of at least one live bug by any method in at least one domicile of a given house compound. The prevalence of house (or domestic) infestation was estimated relative to the total number of houses inspected for infestation. Bug abundance was estimated as the total number of triatomines collected per unit of search effort (0.25 person-hour), as described by Gaspe et al. (2015a).

Vector control personnel sprayed all house compounds with suspension concentrate (SC) deltamethrin (K-Othrin, Bayer) at standard dose (25 mg/m^2) using routine procedures immediately after baseline vector surveys (Gurevitz et al. 2013; Supp. Table 1 [online only]). All bugs sighted while removing furniture and household goods during spraying operations were captured to provide an additional index of house infestation (i.e., during insecticide spraying).

Only houses that were found infested with *T. infestans* during the surveillance phase were sprayed again with SC deltamethrin (as before) or SC beta-cypermethrin (Sipertrin, Chemotecnica, Argentina, at 50 mg/m^2), depending on insecticide availability (Supp. Table 1 [online only]). In some cases, adjacent dwellings were also sprayed owing to their proximity to infested ones. Because of recurrent infestation over three consecutive surveys (following two pyrethroid sprays) and reported vector control failures owing to moderate pyrethroid resistance in Area I of Pampa del Indio (Gurevitz et al. 2012), two houses were sprayed with malathion (Onix, Cheminova, Denmark, at 1 g/m^2) at 21 MPS using procedures described by Gurevitz et al. (2013). Bags and boxes with clothes were not sprayed with malathion. Other houses found to be infested at 39 MPS were sprayed with double-dose SC deltamethrin (K-Othrin, Bayer, Argentina) at 50 mg/m^2 to increase the effectiveness of insecticide treatment (Cecere et al. 2013).

Insect Processing and Diagnosis of *T. cruzi* Infection

All collected bugs were stored in labeled plastic bags and transported to the field laboratory, where they were identified taxonomically and counted according to species, stage, or sex. Only live bugs were examined for *T. cruzi* infection by microscopical observation of fecal samples, as described by Cardinal et al. (2014). The time elapsed between bug collection at baseline and processing was considerably long, and only 82 (19.5%) *T. infestans* captured at four houses from Cancha Larga (i.e., 10.5% of all 38 infested houses) were alive on arrival to the laboratory and could be examined for infection. Infested houses with no bugs examined for infection either had very few insects or bugs were dead by the time they were processed.

Insecticide Resistance

A sample of the *T. infestans* bugs captured at 14, 39, and 51 MPS was tested for pyrethroid (deltamethrin) resistance at the Center for Research on Plagues and Insecticides (CIPEIN/CONICET, Buenos Aires, Argentina) using standard protocols described by Picollo et al. (2005). First-instar nymphs produced by 40 female bugs (including 1-10 insects per house) collected at 10 houses from four communities were tested for resistance (Supp. Table 1 [online only]). Nymphs from each house were pooled, and three independent replicates including 10 insects each were carried out. Nymphal mortality of $<$90% in two out of three replicates was taken as evidence of reduced susceptibility to pyrethroids.

Data Analysis

The data analyzed in this study are included as supplementary files (Supp. Table 2 [online only]).

To identify ecological and sociodemographic determinants of house infestation and bug abundance, we restricted the analyses to domestic habitats because most infestations occurred there. For house compounds with more than one domicile, we used the specific information for each infested domicile (e.g., refuge availability, construction materials), or when all domiciles were not infested, we used the worst condition recorded (e.g., mud walls, oldest premise). For analysis of the relationship between baseline domestic infestation and socioeconomic factors, variables measured over 2009-2012 were back-corrected to 2008 by using the nearest record in time, including variables known to change very little over time (e.g., building material). Most of the missing data were owing to houses left vacant or which ceased to exist rather than by lack of inspection.

Logistic and negative binomial multiple regression analyses were carried out to identify the variables most closely related to baseline domestic infestation and bug abundance, respectively. The models included 166 domiciles. We used a multimodel inference approach based on Akaike's information criterion to estimate the model-averaged effect size (odds ratio, OR) and relative importance (RI) given the variables and models considered (Burnham and Anderson 2002), as described by Gaspe et al. (2015a). The RI of each variable is defined as the sum of Akaike weights in each model in which the variable is present (Burnham and Anderson 2002). We considered the same explanatory variables included in the analog model fitted to baseline data collected in Area III (Gaspe et al. 2015a), except for the goat-equivalent index, which in Area II had many missing data. Housing quality (a three-level categorical variable) was represented by the combination of mud walls (vs. brick-cement walls) and tarred-cardboard sheets on the roof (vs. corrugated-metal sheets).

To assess the predictive performance of the model fitted to baseline infestation in Area III, we cross-validated this model by using the baseline domestic infestation data recorded in Area II and then assessed model fitting by the Hosmer–Lemeshow goodness-of-fit test and the receiver-operating characteristic curve (ROC) implemented in R (version 2.15.1 R Development Core Team 2012), as described by Gaspe et al. (2015a). Multicollinearity was assessed by the variance inflation factor implemented in Stata 12 (Stata Corp 2012, College Station, TX).

Two distinct time periods after community-wide spraying were identified: the early (14–21 MPS) and the late (27–51 MPS) surveillance phase. The former mainly included houses with persistent infestations, whereas the latter comprised infested houses that had not previously been infested after initial spraying. Bivariate risk factor analysis for domestic infestation over these periods was carried out via Firth penalized logistic regression implemented in Stata 12 (Stata Corp 2012, College Station, TX). Firth regression produces finite, consistent estimates of regression parameters when the maximum likelihood estimates do not exist because of complete or quasi-complete data separation and reduces small-sample bias (Heinze and Schemper 2002). The explanatory variables included house infestation at t-1, indoor presence of boxes for storage, distance to the nearest infested house at t-1, and refuge availability. The models included data for 418 houses (early surveillance phase) and 428 houses (late surveillance phase).

Spatial Analysis of House Infestation

The georeferenced location of each house and the infestation data were combined to generate maps of the spatial distribution of *T. infestans* at baseline and postspraying surveys. The spatial pattern of postspraying domestic infestation and bug abundance was assessed by point pattern analysis (PPA) using PPA and Programita software (Wiegand and Moloney 2004, Fortin and Dale 2005). We used the weighted K-function and the random labeling null model to test for spatial aggregation of house and domestic infestation at a global level (Wiegand and Moloney 2004). Given the small number of houses infested on each postspraying survey, all the outcome data from 14 to 51 MPS were pooled. Maximum search distance was 6 km (i.e., one-third of the shortest side of the area, following Fortin and Dale 2005); the search interval was set to 120 m (i.e., the shortest distance between infested houses). We ran 999 Monte-Carlo simulations, and the confidence "envelopes" were calculated with

Table 1. Distribution of house status over time in Area II, Pampa del Indio, 2008–2012, according to whether they were inhabited, inspected, and sprayed during the entomological surveys and their infestation status

House premises	No. of houses (%)					
	0 MPS	14 MPS	21 MPS	27 MPS	39 MPS	51 MPS
Registered	496	501	492	491	492	487
Inhabited	437	421	404	395	391	382
Uninhabited	32	52	60	69	74	79
Public building	27	28	28	27	27	26
Inspected by timed searches	185 (42.3)	381 (90.5)	369 (91.3)	360 (91.1)	337 (86.2)	340 (89.0)
Noninspected[a]	252 (57.7)	40 (9.5)	35 (8.7)	35 (8.9)	54 (13.8)	42 (11.0)
Closed	18 (4.1)	39 (9.3)	24 (5.9)	26 (6.6)	50 (12.8)	39 (10.2)
Rejected	10 (2.3)	1 (0.2)	11 (2.7)	9 (2.3)	4 (1.0)	3 (0.8)
Infested by any method[b]						
Compound	38 (20.5)	19 (5.0)	8 (2.2)	3 (0.8)	14 (4.2)	12 (3.5)
Domicile	27 (14.6)	13 (3.4)	5 (1.4)	2 (0.6)	12 (3.9)	8 (2.4)
Sprayed[c]	409 (93.6)	23[d] (100)	10[d] (100)	3 (100)	12[d] (64.3)[e]	11[d] (75.0)[e]

Only inhabited houses were considered for categories other than registered houses. MPS, months postspraying.

[a] Noninspected houses include those that were closed, rejected inspection, or were not included in the systematic sample.

[b] Number of *T. infestans* collected over subsequent surveys: 420, 465, 53, 9, 106, and 348, respectively. Only houses included in the systematic survey were used for estimating the baseline prevalence of infestation.

[c] The number of sprayed houses over 14–51 MPS includes infested houses and adjacent houses. The percentage of sprayed houses was calculated relative to the number of inhabited houses registered at 0 MPS, and to the number of infested houses registered over 14–51 MPS.

[d] Additional noninfested houses close to infested houses were also sprayed with insecticides.

[e] Unsprayed infested houses include those where householders' notifications were not corroborated by subsequent timed manual searches.

the 25th upper and lower values of all simulations. For assessment of local aggregation of domestic bug abundance (hotspots), we used the Getis statistic (Gi*; Getis and Ord 1996) with the same parameters as the global analysis.

Aggregation at a global level was determined graphically: if the observed values of the K-function at any given distance were higher than that of the confidence "envelopes" generated by Monte-Carlo simulations of the null model, then aggregation of infested houses was considered significant at those distances (Wiegand and Moloney 2004). At a local level, the significance of the Gi* statistic for all distances at any domicile was assessed via the normal approximation, under the null hypothesis that all random permutations were equally likely (Anselin and Rey 2010). A $\log(x + 1)$ transformation was used for bug abundance to reduce overdispersion.

Results

Sociodemographic Profile

In total, 437 inhabited houses, 32 uninhabited houses, and 27 public buildings (including schools, healthcare posts, and churches) were registered at baseline (Table 1). The resident population was composed of Qom (66.6%) and Creole (33.4%) households, whose sociodemographic profiles and house characteristics differed substantially in many respects (Table 2). Qom households were larger and had smaller domiciles (typically made of mud walls and tin roofs) surrounded by fewer peridomestic sites than Creole households. Critical overcrowding (i.e., ≥3 human occupants per sleeping quarter according to INDEC 2010) was almost three times more frequent among Qom (65.5%) than Creole (23.5%) residents. Although most (57.9%) Qom households were engaged in agricultural activities, they had fewer livestock (indexed by goat-equivalents) than Creoles. Seventy-eight percent of the households were on public welfare support. Creole households reported more frequent domestic insecticide use (65.4%) than Qom households (38.8%). The Area II population presented a more heterogeneous sociodemographic profile than Area III (Table 2 in Gaspe et al. 2015a).

House Infestation and Infection With *T. cruzi*

The baseline prevalence of infestation with *T. infestans* was 20.5% for house compounds and 14.6% for domiciles, as determined by any bug collection method (Table 1; Fig. 2). Median domestic bug abundance was relatively low (3 bugs per unit of search effort); most of the bugs were fourth- or fifth-instar nymphs. Peridomestic infestation occurred in 7.6% of house compounds and nearly always comprised ecotopes used at least by chickens: chicken coops, chicken nests, kitchens, and storerooms. Additional consideration of bugs collected during insecticide spraying and by householders increased by 50% the prevalence of house infestation determined by timed manual searches (from 14.1 to 20.5%) and domestic infestation (from 9.7 to 14.6%).

Community-wide spraying with insecticides reached high (93.6%) levels of coverage (Table 1). Rejection levels remained marginal (<3%) throughout the follow-up, whereas the fraction of uninspected houses (mostly owing to being closed) ranged from 8.7 to 13.8%. Only three houses were never assessed for infestation. House infestation rates substantially dropped from 20.5% to 5.0% at 14 MPS, and then fluctuated between 0.8 and 4.2% over 21–51 MPS (Fig. 2; Table 1). Domestic infestation declined similarly from 14.6% at baseline to 2.4% at 51 MPS, as did domestic bug abundance, barring the last observation. Nonetheless, bug abundance and its variability increased at 51 MPS owing to the finding of large bug colonies (>100 bugs) at two houses (Fig. 2). One of the householders had reported the presence of *T. infestans* to the local hospital, but the house was not sprayed. The other household was very close to the former and had relatives living in nearby houses that had been found infested at 39 and 51 MPS. Treatment coverage of houses positive for *T. infestans* between 14 and 51 MPS was also high and ranged from 64.3 to 100% (Table 1). Unsprayed infested houses include those where householders' notification was not corroborated by subsequent timed manual searches. The former index

Table 2. Sociodemographic and housing characteristics according to ethnic group in Area II of Pampa del Indio, Chaco

Variables	Qom (%, No. of houses)	Creole (%, No. of houses)	Total (%, No. of houses)
Sociodemographic characteristics			
Large household size[a]	56.1 (255)	30.7 (127)	47.6 (382)
Critical overcrowding[b]	65.5 (148)	23.5 (68)	52.3 (216)
Insecticide use	38.9 (260)	65.4 (130)	47.7 (390)
Agricultural activities	57.9 (178)	36.9 (103)	50.2 (281)
High goat-equvalent index[c]	29.9 (144)	74.2 (89)	46.8 (233)
Public welfare support	83.1 (172)	69.2 (91)	78.3 (263)
Housing characteristics			
Mud walls	81.3 (262)	41.7 (132)	68.0 (394)
Cardboard roofs	36.2 (260)	11.5 (131)	27.9 (391)
High refuge availability[d]	84.3 (249)	45.8 (131)	71.1 (380)
Recently built[e]	27.9 (79)	24.1 (29)	26.9 (108)
Small domestic area[f]	68.7 (243)	43.4 (122)	60.3 (365)
Electricity	22.5 (217)	58.2 (110)	34.6 (327)
Few or no peridomestic structures[g]	68.2 (286)	42.3 (142)	59.6 (428)

Missing data were excluded from each variable.

[a] More than four people.

[b] Three or more people per bedroom.

[c] More than 30 goat-equivalents.

[d] Includes intermediate and high levels of "Refuge availability".

[e] Less than 5 yr.

[f] Less than 30 m^2 (i.e., median surface of domiciles).

[g] Less than two peridomestic structures (excluding latrines and trees where chickens rested).

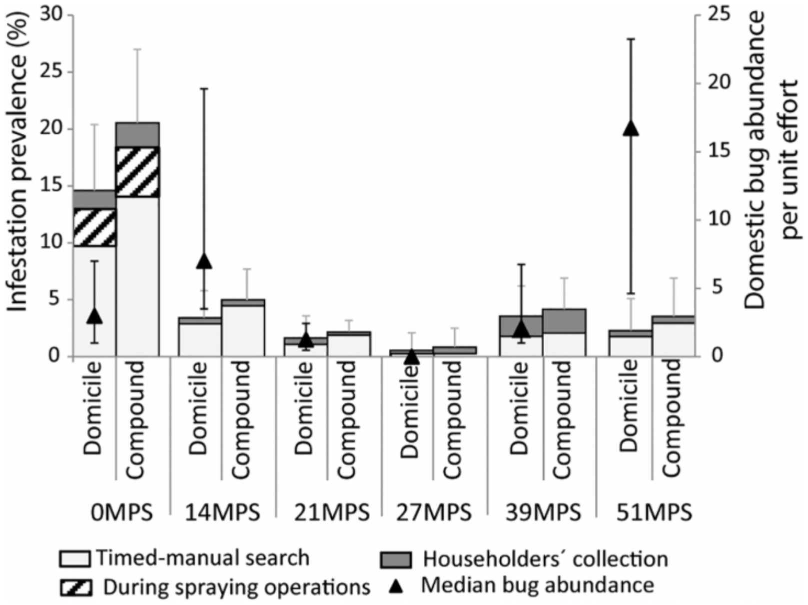

Fig. 2. Prevalence of house and domestic infestation with *T. infestans* and median domestic bug abundance in Area II of Pampa del Indio, Chaco (2008–2012). Infestation was determined by timed manual searches, during spraying and householders' collections. Median bug abundance was calculated for houses infested by timed manual collections. Gray bars, 95% confidence interval for infestation. Black bars, first and third quartiles for domestic bug abundance.

showed poor agreement with house (or domestic) infestation, as determined by any bug collection method (kappa index = 0.2).

Trypanosoma cruzi infection rates in *T. infestans* declined from 41.5% at baseline to 12.4% at 14 MPS, with no infected bug detected among the few examined over 21–27 MPS, and then rose to 1.6 and 10.1% at 39 and 51 MPS, respectively (Table 3). The proportion of houses where at least one bug was examined for infection (among all infested houses) increased to 58.9% over postspraying surveys. Postspraying domestic bug infection rates ranged from 0 to 50% in the few infested houses detected.

Factors Associated With Domestic Infestation

Baseline domestic infestation was closely related to refuge availability (RI = 0.99), housing quality (RI = 0.95), and the occurrence of one or more infested peridomestic structures (RI = 0.91), each of which exerted significant effects ($P < 0.05$) in the multivariate regression model (Table 4). Distance to the nearest infested house, other demographic factors (ethnicity, household size, dogs or cats, chickens), and insecticide use were less important. This model presented a good fit to the data (Hosmer–Lemeshow $\chi^2 = 5.9$, df = 8, $P = 0.66$); the area under the ROC curve was 0.89, with a sensitivity of 88% and specificity of 76%.

Table 3. Occurrence of household infection with *T. cruzi* in domestic or peridomestic *T. infestans* before and after house spraying with insecticides in Area II of Pampa del Indio, Chaco (2008–2012)

Months postspraying (MPS)	% of houses with infected bugs (No. of tested houses[a], No. of infested houses)		% of infected bugs (No. of examined bugs, No. of collected bugs)
	Domicile	House compound	House compound
0	50.0 (2, 27)[b]	25.0 (4, 38)[b]	41.5 (82, 420)
14	50.0 (10, 13)	50.0 (14, 19)	12.4 (185, 465)
21	0.0 (3, 5)	0.0 (6, 8)	0.0 (23, 53)
27	0.0 (0, 2)	0.0 (1, 3)	0.0 (4, 9)
39	14.3 (7, 12)	12.5 (8, 14)	1.6 (62, 106)
51	50.0 (4, 8)	60.0 (5, 12)	10.1 (69, 348)

[a] No of tested houses refers to houses with insects examined for infection.

[b] Infested houses at baseline come out of a systematic survey covering one every three houses.

Table 4. RI and adjusted OR for the multivariate logistic regression model for domestic infestation with *T. infestans* in Areas II ($n = 166$) and III ($n = 386$) of Pampa del Indio, Chaco (2008)

Variable	Area II			Area III		
	RI	OR (CI)	*P*	IR	OR (CI)	*P*
Refuge availability	**0.99**			**1.0**		
Low		1.0			1.0	
Intermediate		4.68 (0.37–59.72)	0.23		2.09 (1.08–4.02)	0.03
High		33.30 (2.36–468.91)	<0.01		4.33 (2.19–8.57)	<0.001
Housing quality[a]	**0.95**			0.19		
1		1.0			1.0	
2		0.14 (0.04–0.52)	<0.01		0.78 (0.44–1.39)	0.40
3		0.51 (0.07–3.72)	0.5		0.68 (0.31–1.53)	0.35
Infested peridomicile	**0.91**			0.66		
Yes		10.9 (1.76–67.09)	0.01		2.41 (0.95–6.10)	0.06
No		1.0			1.0	
Distance to nearest infested house[b]	0.66	0.99 (0.99–1.00)	0.15	**1.0**	0.99 (0.99–0.99)	<0.01
No. of people	0.51	1.14 (0.96–1.36)	0.14	**0.72**	1.07 (1.00–1.15)	<0.05
Ethnic group	0.45			0.49		
Qom		0.33 (0.06–1.93)	0.22		2.27 (0.66–7.86)	0.19
Creole		1.0			1.0	
Presence of poultry	0.45			0.63		
Yes		2.48 (0.66–9.41)	0.18		1.67 (0.95–2.93)	0.07
No		1.0			1.0	
No. of dogs or cats	0.36	1.15 (0.87–1.54)	0.33	0.33	1.04 (0.94–1.16)	0.41
Insecticide use	0.32			**0.74**		
Yes		1.59 (0.53–4.78)	0.41		0.59 (0.35–0.98)	<0.05
No		1.0			1.0	

RI, relative importance; CI, 95% confidence interval.

[a] 1: mud walls and tarred-cardboard sheets on the roof, 2: mud walls and corrugated metal-sheets, 3: brick-cement walls and corrugated metal-sheets.

[b] Per 100 m.

When factors associated with baseline domestic infestation were compared between Areas II and III (Gaspe et al. 2015a), only refuge availability had high RI and large effect size in both areas (Table 4). Despite these differences, the Area III model predicted reasonably well the baseline house infestation status in Area II: the area under the ROC curve was 0.76, with a sensitivity of 79% and specificity of 63%, and a good fit to the data (Hosmer–Lemeshow $\chi^2 = 13.5$; df = 8; $P = 0.1$).

For postspraying domestic infestation over 14–21 and 27–51 MPS, neither of the multivariate models tested was significantly different from the null, and none of the explanatory variables showed significant effects (data not shown). Bivariate analyses indicated that domiciles with higher refuge availability and indoor presence of boxes were at higher risk of being infested at 14–21 and 27–51 MPS, respectively (Table 5).

Spatial Analysis

Most of the infested houses before or after community-wide insecticide spraying were concentrated in the northeastern section of Area II (Fig. 3A and B). However, no global aggregation of postspraying house or domestic infestation was found (Supp. Fig. 1 [online only]). A local spatial analysis of domestic bug abundance detected two hotspots encompassing two infested houses, each within a radius of 1.2 km and 0.25 km, which also included seven to eight noninfested houses (Fig. 3B). Noninfested houses

Table 5. Prevalence of domestic infestation with *T. infestans* and crude OR obtained from bivariate analysis using Firth penalized logistic regression, for variables associated with the main reinfestation hypotheses over 14–21 and 27–51 MPS in Area II of Pampa del Indio, Chaco (2008–2012)

Variable		14–21 MPS		*P*	27–51 MPS		*P*
		% infested[a] (No. of inspected houses)	OR (CI)		% infested[a] (No. of inspected houses)	OR (CI)	
House infestation at t-1[b]							
	Yes	5.4 (37)	2.1 (0.4–10.2)	0.37	4.0 (25)	1.1 (0.2–6.3)	0.89
	No	2.9 (136)	1.0		5.0 (359)	1.0	
Indoor presence of boxes							
	Yes	5.2 (135)	1.8 (0.6–5.0)	0.28	8.7 (127)	2.8 (1.1–6.9)	0.03*
	No	3.0 (234)			3.3 (244)	1.0	
Distance to nearest infested house[c]			1.0 (0.9–1.0)	0.72		1.0 (0.99–1.1)	0.10
	<2.9	3.8 (105)			2.8 (107)		
	2.9–14.4	3.8 (209)			4.7 (214)		
	>14.4	4.8 (104)			6.5 (107)		
Refuge availability							
	Low	0 (124)	1.0		2.9 (136)	1.0	
	Intermediate	2.8 (142)	8.1 (0.4–151.8)	0.16	7.8 (141)	2.59 (0.9–7.9)	0.09
	High	8.8 (148)	24.8 (1.5–421.7)	0.03*	3.4 (145)	1.2 (0.3–4.1)	0.83

[a] Domestic infestation was determined by the finding of at least one live bug by any of the bug collection methods used (i.e., timed manual searches, during insecticide spraying operations, and householders' bug collections).

[b] t-1 refers to baseline house infestation and 14–21 MPS for time periods 14–21 and 27–51 MPS, respectively.

[c] One unit equals 100 m.

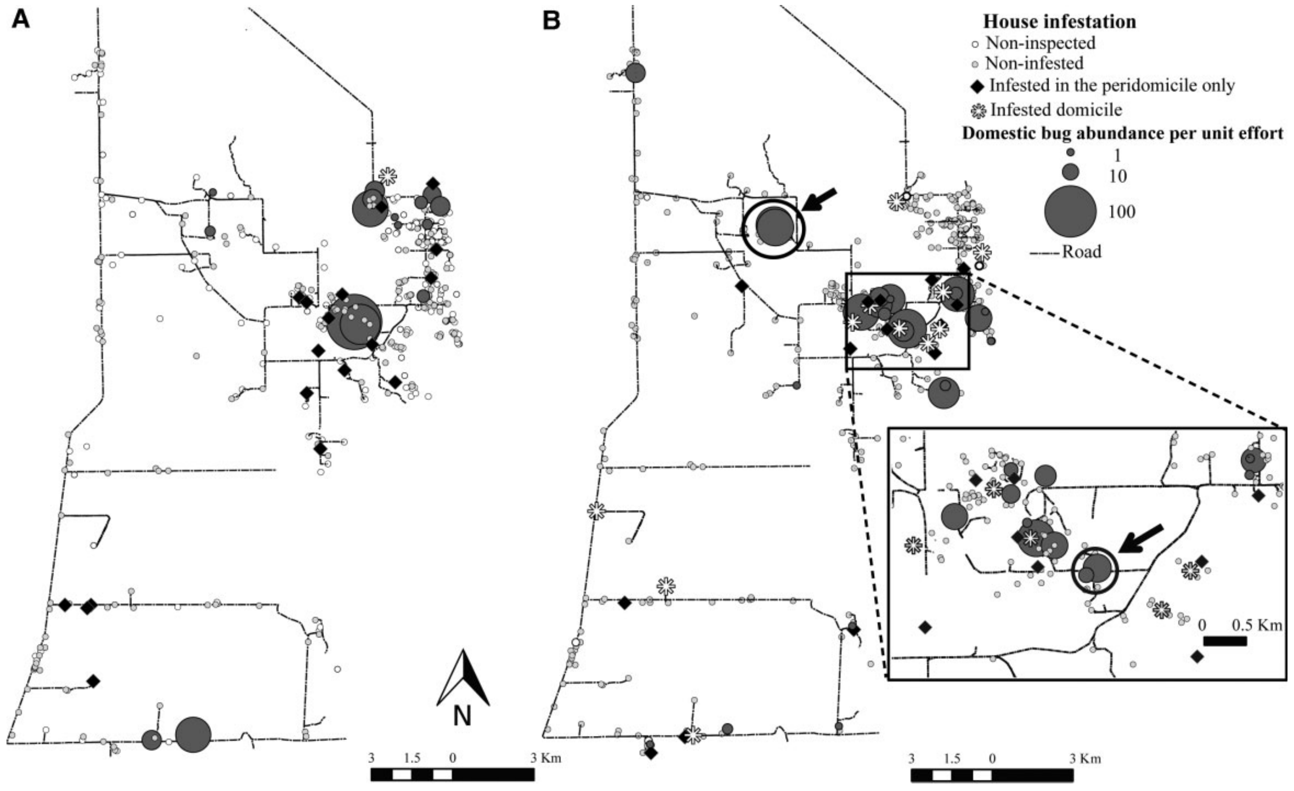

Fig. 3. Baseline (**A**) and postspraying (**B**) house infestation with *T. infestans* and domestic bug abundance in Area II of Pampa del Indio, Chaco (2008–2012). Houses with an infested peridomestic site (black diamonds) were determined by any bug detection method. Houses with an infested domicile (as determined by householders' bug collection or during insecticide spraying) are indicated by a white asterisk. Houses with an infested domicile as determined by timed manual searches are indicated by full gray circles with variable size representing bug abundance per unit effort. Empty circles with a black arrow show the location and extent of infestation hotspots.

within these hotspots were under an increased risk of infestation owing to their proximity to more than one heavily infested house.

Putative Origins of House Reinfestation

We assessed whether postspraying house infestation status was related to prespraying house infestation over the early and late

Table 6. Temporal analysis of house infestation with *T. infestans* status over the early and late surveillance phase in Area II of Pampa del Indio, Chaco (2008–2012)

Prespraying infestation	Postspraying infestation	No. of houses	
		14–21 MPS	27–51 MPS
Yes	Yes	4	3
Yes	No	33	33
No	Yes	4	10
No	No	128	109
Total		169	155

Data include inhabited houses inspected by timed searches at baseline and sprayed with insecticides and then re-inspected during the surveillance phase.

surveillance phase (Table 6). Prespraying house infestation was significantly (exact McNemar's test, $P < 0.001$) and positively associated with postspraying house infestation at 14 MPS (OR = 8.0, 95% CI = 2.8–31.1), over 14–21 MPS (OR = 8.3, 95% confidence interval, 95% CI = 2.9–32.1), and over 27–51 MPS (OR = 3.3, 95% CI = 1.6–7.5). Among the 51 infested houses detected after initial spraying, 30 were not inspected by timed manual searches at baseline (i.e., excluded from the systematic survey or built after baseline). *Triatoma infestans* was detected in seven houses during initial insecticide spraying, four (67%) of which were subsequently infested at 14 MPS.

Pyrethroid resistance bioassays of bugs from two infested houses at 14 MPS yielded reduced bug mortality (62 and 71%) consistent with moderate insecticide resistance; one of these houses was persistently positive in 2006, 2008, and 2009 and was sprayed with insecticide on each occasion. Separate bioassays carried out in bugs from four houses at 39 MPS showed 100% mortality, whereas bugs from other four houses at 51 MPS had mortalities that ranged from 83–87 to 100%. The two houses with a persistent infestation over 0–21 MPS (despite two intervening pyrethroid treatments) sprayed with malathion at 21 MPS were subsequently negative at 27 MPS. However, one of them again harbored a low-density bug colony including adults and fifth-instar nymphs at 39 MPS and was finally suppressed after a double-dose spray with pyrethroids (because malathion had no clearance for use in domestic premises at that time).

A case of putative bug transport (passive or active) from an adjacent municipality under no vector control was detected at 39 MPS. When interviewed about the origins of the single female *T. infestans* collected by timed manual searches, the head of the household reported that her father's premises located 2 km away were infested and family members frequently visited each other and stayed overnight. We subsequently corroborated infestations at this house and in other three nearby houses.

Discussion

Our study documents a sustained impact of a community-wide house spraying with pyrethroids followed by periodic vector surveillance and selective treatments, which reduced the prevalence of house infestation with *T. infestans* from 20.5 to 3.6% over 4 yr after community-wide spraying. Reductions of domestic bug abundance and infection rates up to 27 MPS were even more sizable and compatible with marginal domestic transmission risks (Gürtler et al. 2007, Cardinal et al. 2014), with point occurrences of high bug abundance at 51 MPS. The space–time distribution of house reinfestation suggested the occurrence of two main processes over two successive stages: the first one apparently derived from internal sources that generated persistent house infestations up to 21 MPS (partly associated with moderate pyrethroid resistance), and the second one involving new foci from mostly unidentified sources of *T. infestans* (external or internal to the study area) that arrived via passive or active bug transport over 27–51 MPS. Over both stages, houses found to be infested before blanket insecticide spraying had an increased probability of harboring a subsequent infestation, suggesting the occurrence of stable determinants of house infestation related to precarious construction, vector control practices, and relative location (Gürtler and Yadon 2015).

Baseline house infestation (20.5%) was substantially lower than in the other two rural areas (31.9–45.9%) of Pampa del Indio (Gurevitz et al. 2011, Gaspe et al. 2015a) and elsewhere in the dry Argentine Chaco, as determined by the same methods 6-12 yr after the last insecticide spraying campaign (Gürtler et al. 2004, 2007; Cecere et al. 2006, 2013; Porcasi et al. 2006). The lower infestation levels recorded in Area II were in part related to the insecticide applications carried out 2 yr before our interventions, better housing construction, and other less suitable conditions for *T. infestans*. In comparison with Area III (Gaspe et al. 2015a), Area II houses mainly had tin roofs rather than tarred-cardboard roofs, fewer or no peridomestic structures, smaller household size, greater wealth (indexed by the goat-equivalent index), and more houses with electricity. Multimodel inference analysis corroborated the relevance of precarious housing structure and refuge availability to domestic infestation and abundance of *T. infestans* in the three rural areas of Pampa del Indio and elsewhere in the Gran Chaco (Cecere et al. 1998, Gurevitz et al. 2011, Saunders et al. 2012, Gaspe et al. 2015a). Other risk factors not included in our analyses may contribute to the observed differences among areas.

The community-wide insecticide spraying campaign in Area II exerted relatively larger (Area I) or smaller (Area III) impacts on baseline house infestation and domestic bug abundance under similarly high levels (≥93.6%) of treatment coverage (Gurevitz et al. 2013, Gaspe et al. 2015b). Likewise in Area I, some *T. infestans* populations collected at 14 MPS had moderate pyrethroid resistance that led to apparent vector control failures and residual foci (Gurevitz et al. 2012). These resistance levels were far below those recorded in *T. infestans* elsewhere in northwestern Argentina and Bolivia (Picollo et al. 2005, Lardeux et al. 2010, Mougabure-Cueto and Picollo 2015), including highly resistant foci detected near Pampa del Indio (Carvajal et al. 2012). However, the limited number of parental female bugs collected for screening tests in Area II may provide a biased estimate of the average degree of pyrethroid resistance (Amelotti et al. 2011). The persistent bug colonies detected over 14-21 MPS were finally suppressed after repeated selective sprays with pyrethroids or malathion, thus increasing operational expense and householders' complaints to the unpleasant smell of malathion.

Triatomine persistence at house level was also apparently related to technical failures during insecticide applications in at least one case: lack of mobilization of large boxes for clothing storage and furniture for malathion spraying at 21 MPS was followed by the finding of a large bug colony at 39 MPS. Although we cannot rule out a flight-mediated invasion from a heavily infested house located at 1.5 km, similar operational problems involving partial treatment coverage were documented elsewhere in the Argentine Chaco (Cecere et al. 1997, Gurevitz et al. 2012) and substantially compromise the effectiveness of insecticide treatments. Consistent with these observations, one of the factors significantly associated with postspraying infestation over the late surveillance phase was the occurrence of boxes for clothing storage that frequently had signs of little use. These boxes frequently served as egg-laying sites and as refuges where triatomines may escape from the insecticide spray or hide until its residual effects wane. If not handled properly and treated with insecticides following good practices of bug control, boxes for clothing storage are likely to generate residual foci.

A second stage during the surveillance phase, spanning over 27–51 MPS, was signaled by the finding of triatomines mostly in houses that had not been infested over two or more prior surveys. These new infestations were likely generated by active or passive bug dispersal from internal or external foci. External sources were illustrated by a putative case of bug transport from an infested rural household off the southern border of Pampa del Indio, but there were other infested houses at 2 km of linear distance from which flight dispersal might have occurred. Thus, border areas under no effective vector control may act as sources of house reinfestation with *T. infestans* and jeopardize local elimination attempts. Although sylvatic foci of *T. infestans* were not detected in Areas I and II (Alvarado-Otegui et al. 2012; Y. M. P., unpublished data), its occurrence cannot be definitely excluded. Other potential sources of residual foci involving closed, vacant, or nonparticipating households were very unlikely in light of repeated bug searches and considerable efforts to achieve full-coverage vector assessments and spray operations throughout the follow-up.

Spatial analyses revealed two hotspots of domestic infestation in Qom sections that coincided with persistent foci and evidence of local, moderate pyrethroid resistance at least in two houses. Its aggregation distance (0.25 and 1.2 km) fell within the flight range (2.4 km) of *T. infestans* inferred from the duration of sustained tethered flights and spatial patterns of house reinfestation (Cecere et al. 2006, Gurevitz et al. 2006), whereas the observed flight range of marked adult bugs released in a salt flat likely exceeded 550 m (Schofield et al. 1992). Thus, the observed hotspots may have been generated by bug dispersal from infested houses. Most Area II houses were vulnerable to intrusive triatomines because of their precarious physical structure, lack of window screens, and proximity to other houses. The association between prespraying and postspraying infestation over 14–21 and 27–51 MPS also indicates that some houses were at increased risks of infestation regardless of the precise pathway by which bugs invaded the house.

Analysis of household mobility and residence patterns in Area II does not support the supposition that they played a significant role as a risk factor for house infestation as in Area III. Despite that most of the Area II households were of Qom descent, their much lower frequency of recently built houses suggests less intense mobility than in Area III, where Qom households frequently dismantled and rebuilt their houses within the same rural section (Gaspe et al. 2015a). A fraction of Creole (19%) and Qom (5%) households reportedly owned and partially occupied another house at the local town, thus revealing a steady flow of people and goods between areas and potential threats of bug transport in either direction (Provecho et al. 2014). A note of caution is needed because the precision of self-reported variables is affected by recall bias. Creoles comprised one-third of households and their houses were, on average, less frequently infested than Qom's at baseline.

Determining whether an infestation is persistent or not is affected by the sensitivity of bug detection methods, time elapsed between vector surveys, and context. Timed manual searches (with or without a dislodging aerosol) underestimate the true house infestation rates when bug density is low, especially after insecticide spraying (Gürtler et al. 1995, Abad-Franch et al. 2011, Gurevitz et al. 2012, Barbu et al. 2014). These studies also showed that in systematic searches during insecticide spraying operations (including knockdown bug collections over a few days posttreatment), householders frequently detected infestations missed by timed manual searches. Our baseline survey corroborated that approximately 50% of all house infestations were undetected by timed manual searches. Moreover, householders' notifications of the domestic presence of *T. infestans* (with no bug kept) had a poor agreement with timed manual searches as in Area III (Gaspe et al. 2015a) and were not corroborated by subsequent timed searches. When insecticide treatments strongly reduce bug abundance below the detection threshold of timed manual searches, "false negative" results are more likely than over subsequent occasions following recovery of bug population size. Thus, a time series of house infestation data before and shortly after insecticide spraying may show a negative finding in between two positive searches spaced wide apart in time. Distinguishing between locally persistent infestations and new establishments generated by bugs invading from elsewhere is challenging and microsatellite or morphometric markers may provide additional evidence (Peréz de Rosas et al. 2008, Piccinali and Gürtler 2015, Gaspe et al. 2015b). In the current study, because the full-coverage insecticidal campaign strongly curtailed the number of bug sources that may fuel house recolonization events, houses infested both at 0 and 14–21 MPS most likely were persistent foci that survived one or two insecticide applications rather than new foci.

Our study has some limitations. Spatial and temporal analyses of house infestation data were limited by lack of full-coverage inspections at baseline, which reduced the sample size of sites examined both at baseline and 14–21 or 27–51 MPS. To relate the occurrence of postintervention domestic infestations to putative risk factors, we pooled the limited number of reinfested houses within each of the time periods, but the absolute frequency of infested houses was small. The spatial analysis was limited by the small number of reinfested houses, and therefore, we only explored the general patterns of reinfestation. The overdispersed distribution of bug abundance also limited local spatial analysis despite using a log transformation. Nonetheless, the same qualitative results were obtained with and without the transformation. Baseline bug infection rates were based on very few houses. A particular strength of the current research effort is the large number of georeferenced households surveyed repeatedly over 4 yr and a detailed examination of the various hypotheses on the putative origins of postspraying bugs.

The diminished effectiveness of single insecticidal treatments, partly attributable to moderate pyrethroid resistance and further compounded by the limited sensitivity of timed manual searches, contributed to the persistence of *T. infestans* in Pampa del Indio, albeit at lower abundance levels. Poor housing facilitated house recolonization by triatomines invading from various sources, and high rates of human infection with *T. cruzi* explain the fast emergence of domestic bug infection shortly after houses were recolonized. The multiple factors and sources of house reinfestation operating after community-wide and selective insecticide spraying suggest the need to implement sustained, integrated control strategies with coordinated spatial coverage to minimize the risk of vector introduction or invasion from peripheral, uncontrolled areas. Householders' active participation in the early detection of new infestations is essential to prevent house recolonization, bug population recovery, and subsequent propagation. A crucial requisite for effective vector surveillance is the rapid response (insecticide spray) of the local health system to householders' bug notifications. Strengthening the local response component through inclusion of qualified vector control personnel and provision of the necessary gear is in point. Improved disease and vector control strategies combined with broad social participation are needed for the sustainable elimination of vector-borne human infection with *T. cruzi* from the Gran Chaco.

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Supplementary Data

Supplementary data are available at *Journal of Medical Entomology* online.

References

Abad-Franch, F., M. C. Vega, M. S. Rolón, W. S. Santos, and A. Rojas de Arias. 2011. Community participation in Chagas disease vector surveillance: Systematic review. PLoS Negl. Trop. Dis. 5: e1207.

Abrahan, L. B., D. E. Gorla, and S. S. Catalá. 2011. Dispersal of *Triatoma infestans* and other Triatominae species in the arid Chaco of Argentina: flying, walking or passive carriage? The importance of walking females. Mem. Inst. Oswaldo Cruz 106: 232–239.

Alvarado-Otegui, J. A., L. A. Ceballos, M. M. Orozco, G. F. Enriquez, M. V. Cardinal, C. Cura, A. G. Schijman, U. Kitron, and R. E. Gürtler. 2012. The sylvatic transmission cycle of *Trypanosoma cruzi* in a rural area in the humid Chaco of Argentina. Acta Trop. 124: 79–86.

Amelotti, I., N. Romero, S. S. Catalá, and D. E. Gorla. 2011. Variability of the susceptibility to deltamethrin in *Triatoma infestans* (Hemiptera: Reduviidae). The female factor. J. Med. Entomol. 48: 1167–1173.

Anselin, L., and S. J. Rey. 2010. Perspectives on spatial data analysis. Springer-Verlag.

Bacigalupo, A., F. Torres-Pérez, V. Segovia, A. García, J. P. Correa, L. Moreno, P. Arroyo, and P. E. Cattan. 2010. Sylvatic foci of the Chagas disease vector *Triatoma infestans* in Chile: Description of a new focus and challenges for control programs. Mem. Inst. Oswaldo Cruz 105: 633–641.

Barbu, C., A. Buttenheim, M. Hancco Pumahuanca, J. E. Quintanilla Calderón, R. Salazar, M. Carrión, A. Catacora Rospigliossi, F. S. Malaga Chavez, K. Oppe Alvarez, J. Cornejo del Carpio, et al. 2014. Residual infestation and recolonization in an urban *Triatoma infestans* control campaign, Peru. Emerg. Infect. Dis. 20: 2055–2063.

Bayer, A. M., G. C. Hunter, R. H. Gilman, J. G. Cornejo del Carpio, C. Naquira, C. Bern, and M. Z. Levy. 2009. Chagas disease, migration and community settlement patterns in Arequipa, Peru. PLoS Negl. Trop. Dis. 3: e567.

Brenière, S. F., R. Salas, R. Buitrago, P. Brémond, V. Sosa, M. F. Bosseno, E. Waleckx, S. Depickère, and C. Barnabé. 2013. Wild populations of *Triatoma infestans* are highly connected to intra peridomestic conspecific populations in the Bolivian Andes. PLoS ONE 8: e80786.

Briceño-León,, R., and J., Méndez Galván. 2007. The social determinants of Chagas disease and the transformations of Latin America. Mem. Inst. Oswaldo Cruz 102: 109–112.

Burnham, K.P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach. Springer-Verlag.

Bustamante, D. M., S. M. De Urioste-Stone, J. G. Juárez, and P. M. Pennington. 2014. Ecological, social and biological risk factors for continued *Trypanosoma cruzi* transmission by *Triatoma dimidiata* in Guatemala. PLoS ONE 9: e104599.

Cardinal, M. V., M. M. Orozco, G. F. Enriquez, L. A. Ceballos, M. S. Gaspe, J. A. Alvarado-Otegui, J. M. Gurevitz, U. Kitron, and R. E. Gürtler. 2014. Heterogeneities in the ecoepidemiology of *Trypanosoma cruzi* infection in rural communities of the Argentinean Chaco. Am. J. Trop. Med. Hyg. 90: 1063–1073.

Carvajal, G., G. Mougabure-Cueto, and A. C. Toloza. 2012. Toxicity of non-pyrethroid insecticides against *Triatoma infestans* (Hemiptera: Reduviidae). Mem. Inst. Oswaldo Cruz 107: 675–679.

Ceballos, L. A., R. V. Piccinali, I. Berkunsky, U. Kitron, and R. E. Gürtler. 2009. First finding of melanic sylvatic *Triatoma infestans* (Hemiptera: Reduviidae) colonies in the Argentine Chaco. J. Med. Entomol. 46: 1195–1202.

Cecere, M. C., R. E. Gürtler, D. Canale, R. Chuit, and J. E. Cohen. 1997. The role of peridomiciliary area in the elimination of *Triatoma infestans* from rural Argentine communities. Pan. Am. J. Public Health 1: 273–279.

Cecere, M. C., R. E. Gürtler, R. Chuit, and J. E. Cohen. 1998. Factors limiting the domestic density of *Triatoma infestans* in north-west Argentina: A longitudinal study. Bull. World Health Org. 76: 373–384.

Cecere, M. C., G. M. Vazquez-Prokopec, L. A. Ceballos, J. M. Gurevitz, J. E. Zarate, M. Zaidenberg, U. Kitron, and R. E. Gürtler. 2006. Comparative trial of effectiveness of pyrethroid insecticides against peridomestic populations of *Triatoma infestans* in northwestern Argentina. J. Med. Entomol. 43: 902–909.

Cecere, M. C., G. M. Vázquez-Prokopec, L. A. Ceballos, S. Boragno, J. E. Zárate, U. Kitron, and R. E. Gürtler. 2013. Improved chemical control of Chagas disease vectors in the dry Chaco region. J. Med. Entomol. 50: 394–403.

Dias, J.C.P., and R. B. Dias. 1982. Housing and the control of vectors of human Chagas' disease in the State of Minas Gerais, Brazil. Rev. Panam. Salud Pública. 16: 117–129.

Dias, J.C.P., A. C. Silveira, and C. J. Schofield. 2002. The impact of Chagas disease control in Latin America. Mem. Inst. Oswaldo Cruz 97: 603–612.

Dumonteil, E., P. Nouvellet, K. Rosecrans, M. J. Ramírez-Sierra, R. Gamboa-León, V. Cruz-Chan, M. Rosado-Vallado, and S. Goubière. 2013. Eco-bio-social determinants for house infestation by non-domiciliated *Triatoma dimidiata* in the Yucatán Península, México. PLoS Negl. Trop. Dis. 7: e2466.

Fortin, M. J., and M.R.T. Dale. 2005. Spatial analysis: A guide for ecologists. Cambridge University Press.

Gaspe, M. S., Y. M. Provecho, M. V. Cardinal, M. P. Fernández, and R. E. Gürtler. 2015a. Ecological and sociodemographic determinants of house infestation by *Triatoma infestans* in indigenous communities of the Argentine Chaco. PLoS Negl. Trop. Dis. 9: e0003614.

Gaspe, M. S., Y. M. Provecho, R. V. Piccinali, and R. E. Gürtler. 2015b. Where do these bugs come from? Phenotypic structure of *Triatoma infestans* populations after control interventions in the Argentine Chaco. Mem. Inst. Oswaldo Cruz 110: 310–318.

Getis, A., and J. K. Ord. 1996. Local spatial statistics: an overview. *In* P. Longley, M. Batty (eds), Spatial analysis modeling in a GIS environment. GeoInformation International, pp. 269–285.

Gurevitz, J. M., L. A. Ceballos, U. Kitron, and R. E. Gürtler. 2006. Flight initiation of *Triatoma infestans* (Hemiptera: Reduviidae) under natural climatic conditions. J. Med. Entomol. 43: 143–150.

Gurevitz, J. M., L. A. Ceballos, M. S. Gaspe, J. A. Alvarado-Otegui, G. F. Enriquez, U. Kitron, and R. E. Gürtler. 2011. Factors affecting infestation by *Triatoma infestans* in a rural area of the humid Chaco in Argentina: A multimodel inference approach. PLoS Negl. Trop. Dis. 5: e1365.

Gurevitz, J. M., M. S. Gaspe, G. F. Enríquez, C. V. Vassena, J. A. Alvarado-Otegui, Y. M. Provecho, G. A. Mougabure Cueto, M. I. Picollo, U. Kitron, and R. E. Gürtler. 2012. Unsuspected pyrethroid resistance and control failures of Chagas disease vector in Argentina. J. Med. Entomol. 49: 1379–1386.

Gurevitz, J. M., M. S. Gaspe, G. F. Enriquez, Y. M. Provecho, U. Kitron, and R. E. Gürtler. 2013. Intensified surveillance and insecticide-based control of the Chagas disease vector *Triatoma infestans* in the Argentinean Chaco. PLoS Negl. Trop. Dis. 7: e2158.

Gürtler, R. E. 2009. Sustainability of vector control strategies in the Gran Chaco Region: Current challenges and possible approaches. Mem. Inst. Oswaldo Cruz 104: 52–59.

Gürtler, R. E., and Z. Yadon. 2015. Eco-bio-social research on community-based approaches for Chagas disease vector control in Latin America. Trans. R. Soc. Trop. Med. Hyg. 109: 91–98.

Gürtler, R. E., R. Chuit, M. C. Cecere, and M. B. Castañera. 1995. Detecting domestic vectors of Chagas disease: A comparative trial of six methods in north-west Argentina. Bull. World Health Organ. 73: 487–494.

Gürtler, R. E., D. M. Canale, C. Spillmann, R. Stariolo, O. D. Salomon, S. Blanco, and E. L. Segura. 2004. Effectiveness of residual spraying of peridomestic ecotopes with deltamethrin and permethrin on *Triatoma infestans* in rural western Argentina: A district-wide randomized trial. Bull. World Health Organ. 82: 196–205.

Gürtler, R. E., U. Kitron, M. C. Cecere, E. L. Segura, and J. E. Cohen. 2007. Sustainable vector control and management of Chagas disease in the Gran Chaco, Argentina. Proc. Natl. Acad. Sci. 104: 16194–16199.

(INDEC) Instituto Nacional de Estadísticas y Censos 2010. Censo nacional de población, hogares y viviendas. (http://www.censo2010.indec.gov.ar/, accessed 7 January 2016)

Heinze, G., and M. Schemper. 2002. A solution to the problem of separation in logistic regression. Statistics in Medicine. 21: 2409–19.

Hotez, P. J., M. E. Botazzi, C. Franco-Paredes, S. K. Ault, and M. R. Periago. 2008. The neglected tropical diseases of Latin America and the Caribbean: a review of disease burden and distribution and a roadmap for control and elimination. PLoS Negl. Trop. Dis. 2: e300.

Lardeux, F., S. Depickere, S. Duchon, and T. Chavez. 2010. Insecticide resistance of *Triatoma infestans* (Hemiptera, Reduviidae) vector of Chagas disease in Bolivia. Trop. Med. Int. Health 15: 1037–1048.

Lee, B. Y., K. M. Bacon, M. E. Bottazzi, and P. J. Hotez. 2013. Global economic burden of Chagas disease: A computational simulation model. Lancet Infect Dis. 13: 342–348.

Ministry of Agriculture 2010. Ganados y carnes. 2010 (cited 2014 May 8). p. 452. (http://www.minagri.gob.ar/site/ganaderia/anuario/pdf2010/ANUARIO 2010 COMPLETO web.pdf, accessed 8 July 2015)

Mougabure-Cueto, G., and M. I. Picollo. 2015. Insecticide resistance in vector Chagas disease: Evolution, mechanisms and management. Acta Trop. 149: 70–85.

Noireau, F., M. G. Rojas Cortez, F. A. Monteiro, A. M. Jansen, and F. Torrico. 2005. Can wild *Triatoma infestans* foci in Bolivia jeopardize Chagas disease control efforts? Trends Parasitol. 21: 12.

Peréz de Rosas, A. R., E. L. Segura, L. Fichera, and B. A. García. 2008. Macrogeographic and microgeographic genetic structure of the Chagas' disease vector *Triatoma infestans* (Hemiptera: Reduviidae) from Catamarca, Argentina. Genetica 133: 247–260.

Piccinali, R. V., and R. E. Gürtler. 2015. Fine-scale genetic structure of *Triatoma infestans* in the Argentine Chaco. Inf. Gen. Evol. 34: 143–152.

Picollo, M. I., C. Vassena, P. S. Orihuela, S. Barrios, M. Zaidemberg, and E. Zerba. 2005. High resistance to pyrethroid insecticides associated with ineffective field treatments in *Triatoma infestans* (Hemiptera: Reduviidae) from northern Argentina. J. Med. Entomol. 42: 637–642.

Porcasi, X., S. S. Catalá, H. Hrellac, M. C. Scavuzzo, and D. E. Gorla. 2006. Infestation of rural houses by *Triatoma infestans* (Hemiptera: Reduviidae) in southern area of Gran Chaco in Argentina. J. Med. Entomol. 43: 1060–1067.

Provecho, Y. M., M. S. Gaspe, M. P. Fernández, G. F. Enriquez, D. Weinberg, and R. E. Gürtler. 2014. The peri-urban interface and house infestation with *Triatoma infestans* in the Argentine Chaco: An underreported process? Mem. Inst. Oswaldo Cruz 109: 923–934.

R Development Core Team 2012. R: A language and environment for statistical computing. Release 2.15.1. Vienna: R Foundation for Statistical Computing.

Rolón, M., M. C. Vega, F. Román, A. Gómez, and A. R. de Arias. 2011. First report of colonies of sylvatic *Triatoma infestans* (Hemiptera: Reduviidae) in the Paraguayan Chaco, using a trained dog. PLoS Negl. Trop. Dis. 5: e1026.

Saunders, M., A. Small, M. Dedicoat, and L. Roberts. 2012. The development and validation of a risk score for household infestation by *Triatoma infestans*, a Bolivian vector of Chagas disease. Trans. R. Soc. Trop. Med. Hyg. 160: 677–682.

Schmunis, A. G., and Z. E. Yadón. 2010. Chagas disease: A Latin American health problem becoming a world health problem. Acta Trop. 110: 14–21.

Schofield, C. J., M. J. Lehane, P. McEwen, S. S. Catalá, and D. E. Gorla. 1992. Dispersive flight by *Triatoma infestans* under natural climatic conditions in Argentina. Med. Vet. Entomol. 6: 51–56.

Schofield, C. F., J. Jannin, and R. Salvatella. 2006. The future of Chagas disease control. Trends Parasitol. 22: 583–588.

Stata Corp 2012. Stata Statistical Software. Release 12.0. Stata Corporation, College Station.

Vázquez-Prokopec, G. M., L. A. Ceballos, P. L. Marcet, M. C. Cecere, M. V. Cardinal, U. Kitron, and R. E. Gürtler. 2006. Seasonal variations in active dispersal of natural populations of *Triatoma infestans* in rural north-western Argentina. Med. Vet. Entomol. 20: 1–6.

(WHO) World Health Organization 2002. Control of Chagas disease: Second report of the WHO expert committee. World Health Organization, Geneva.

(WHO) World Health Organization 2015. Weekly Epidemiological Record 90: 33–44.

Wiegand, T., and K. A. Moloney. 2004. Rings, circles and null-models for point pattern analysis in ecology. Oikos 104: 209–229.