Deltamethrin-impregnated bednets reduce human landing rates of sandfly vector *Lutzomyia longipalpis* in Amazon households

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**Abstract.** The entomological efficacy of using 25% deltamethrin EC insecticide-treated bednets (ITNs) was evaluated against the sandfly *Lutzomyia longipalpis* Lutz and Neiva (Diptera: Psychodidae), the principal vector of zoonotic visceral leishmaniasis (ZVL) in Latin America. A crossover field study in Amazon Brazil (Marajó Island, Pará State) demonstrated that, compared with untreated nets, the insecticide increased the barrier effect of nets by 39% (95% confidence interval [CI] 34–44%), reduced human landing rates by 80% (95% CI 62–90%) and increased the 24-h mortality rate from 0% to 98% (95% CI 93–99%) inside ITNs. The presence of an ITN also reduced the human landing rate on unprotected persons outside the net in the same room by 56% (95% CI 52–59%), and increased 24-h mortality to 68% (95% CI 62–73%) compared to 0.4% (0.1–2.0%) in untreated houses. The reduction in human landing rates in ITN rooms was associated with a doubling in the proportion of sandflies alighting on walls compared with that in untreated rooms, which was attributed to insecticide-induced excito-repellency. There was no evidence that sandflies were diverted onto unprotected hosts. Human landing catches inside houses peaked between 19.00 hours and 23.00 hours and declined steadily to zero at 02.00 hours and thereafter. House-to-house questionnaires established that only 34% of households owned at least one net (median two, range 1–8), only 20% of the population slept under a net (33% of 0–5-year-old children), and the majority (73%) of the population slept in hammocks. Combined data pertaining to sleeping times for children and sandfly activity period indicate that >50% of sandfly bites inside houses, and substantially more outside houses, were taken before a third of children were potentially protected by a net. This study demonstrates the clear entomological efficacy of ITNs against *Lu. longipalpis* in this endemic region. The effectiveness of ITNs at preventing ZVL infection and disease has still to be evaluated.

**Key words.** *Lutzomyia longipalpis*, bednets, insecticide, ITNs, leishmaniasis, sandfly, vector control, Brazil.

**Introduction**

Insecticide-treated bednets (ITNs) have been highly successful in protecting humans against bites of nuisance *Culex* and *Mansonia* mosquitoes (Lindsay & Gibson, 1988), and arthropod vectors of malaria, lymphatic filariasis, Chagas disease and leishmaniasis (Bogh et al., 1998; Kroeger et al., 1999b; Alexander & Maroli, 2003; Lengeler, 2004), notably resulting in reductions in childhood malaria morbidity and mortality (Lengeler, 2004). Simulating a human-baited trap, the success...
of ITNs is primarily determined by the propensity of arthropod vectors to feed on humans at the time (usually crepuscular/night times) and place (peri-domestic, usually indoors) when humans are protected under a net. By contrast, the efficacy of ITNs against more exophagic, exophilic and zoophilic vectors is less certain (Kroeger et al., 1999a), and interpopulation variation in biting behaviour, in some cases the consequence of short- or longer-term exposure to ITNs (reviewed by Takken, 2002), highlights the requirement to understand the entomological effects of ITNs in any particular epidemiological setting.

Measurable entomological outcomes following exposure to ITNs inside houses include (a) changes in the human landing rate, feeding success and human blood index (HBI); (b) increased vector mortality (measured as immediate ‘knockdown’ and delayed 24-h mortality), mass reduction in vectors in treated areas and lower vector survival (i.e. lower parous rates); (c) excito-repellency (irritation and disorientation) resulting in reduced values in (a) and induced exophily, and (d) deterrency from contact with insecticide-treated surfaces resulting in reduced endophily. Whereas effect (b) is associated with prolonged contact with treated surfaces, (c) is associated with brief (non-lethal) contact with insecticide-treated surfaces, and (d) is usually caused by volatiles contained within the insecticide formulation or contaminated air-borne dust from treated unplastered house walls (Lindsay et al., 1991; Chandre et al., 2000; Maxwell et al., 2002). All else being equal, vectorial capacity is most sensitive to changes in values of parameters in (a) and (b), where humans are the reservoir. High coverage with ITNs may result in reduced vector infection rates (i.e. a mass effect) in addition to providing personal protection against infective bites. Mass effects of ITN intervention against zoonotic organisms (where humans are not reservoirs) are limited to impacting on vector abundance and survival. Under either scenario, it is necessary to ensure that effects (c) and (d) do not inadvertently divert potentially infectious bites away from protected hosts and onto unprotected hosts (Lines et al., 1987).

Field experiments have shown ITNs to reduce human landing rates of sandflies, including cutaneous leishmaniasis vectors Lutzomyia youngi Felicianelli and Murillo in Colombia (Alexander et al., 1995) and Lutzomyia ovallesi Ortiz in Venezuela (Felicianelli et al., 1995), and to reduce the abundance of Lu. youngi and Lu. ovallesi in Venezuela (Kroeger et al., 2002). The efficacy of ITNs against leishmaniasis has been demonstrated by a handful of community-based trials focused on Old World phlebotomine sandfly species, principally Phlebotomus sergenti Parrot, vector of Leishmania tropica (Wright) (Kinetoplastida: Trypanosomatidae) in peri-domestic/urban foci (Tayeh et al., 1997; Reyburn et al., 2000; Alten et al., 2003; Yaghoobi-Ershadi et al., 2006; Jalouk et al., 2007). The only reported efficacy trial against New World leishmaniasis targeted Lu. youngi and Lu. ovallesi in Trujillo, Venezuela, resulting in a 100% reduction in the incidence of human cutaneous lesions in households fitted with insecticide-impregnated curtains compared with an average 4% increase in incidence in control houses (Kroeger et al., 2002).

The more immediate life-threatening form of human leishmaniasis amongst the immunocompetent population is anthroponotic visceral leishmaniasis (AVL) due to Leishmania donovani (Laveran and Mesnil) in the Indian subcontinent and parts of central Asia and Africa, and zoonotic visceral leishmaniasis (ZVL) due to Leishmania infantum Cunha and Chagas (= Leishmania chagasi) in Latin America. Both forms are usually fatal in untreated or unresponsive patients. Case-control and prospective cohort studies of AVL in Nepal and Bangladesh report that use of untreated bednets was associated with a 30–70% lower risk of developing the disease (Bern et al., 2000, 2005) and ITN field trials against AVL are currently ongoing in India and Nepal. The principal vector in the Indian subcontinent is Phlebotomus argentipes Annandale and Brunetti, which is an opportunistic feeder preferring accessible larger hosts such as bovids, to the extent that cattle ownership has proven zooprophylactic effects (Bern et al., 2000, 2005).

No studies have yet assessed the effects of ITNs against vectors of ZVL in Latin America, where Lutzomyia longipalpis Lutz and Neiva is the principal vector. Lutzomyia longipalpis is predominantly peridomestic and, like Ph. argentipes, has catholic feeding habits (Quinnell et al., 1992). In this continent ZVL is a major public health problem, particularly amongst poorer people and malnourished children; domestic dogs are the important reservoir host throughout the range of Le. infantum (Courtenay et al., 2002a; Gavagni et al., 2002a). Widespread vector control activities against ZVL in Brazil are limited to household spraying with insecticides (mainly cypermethrin or deltamethrin), but coverage is sporadic and incomplete due to operational difficulties.

Here we report on a crossover field study carried out in an endemic region of Amazon Brazil to evaluate the entomological efficacy of ITNs for personal protection against Lu. longipalpis, and provide data on sandfly periodicity relative to community use of sleeping nets.

**Materials and methods**

**Study site**

The study was conducted between July and August 2003 (dry season) in the community of Pingo d’Água, municipality of Salvaterra, Marajó Island, Pará State, Brazil (48°03’ W, 00°46’ S), where ZVL is endemic and the behavioural ecology of Lu. longipalpis has been studied extensively (e.g. Lainson et al., 1990; Dye et al., 1991; Quinnell et al., 1992; Quinnell & Dye, 1994a, 1994b; Kelly et al., 1997; Kelly & Dye, 1997).

**Study house recruitment and sandfly activity period**

Indoor household sandfly abundance was assessed by setting a CDC light trap (John W. Hock Co., Gainesville, FL, U.S.A.) on three consecutive nights in the bedroom (night 1), kitchen (night 2) and living room (night 3) of seven houses between 18.00 hours and 06.00 hours. The houses with the highest geometric mean number of female sandflies were enrolled into the crossover study. In the two houses with the highest mean counts, hourly human landing catches were monitored between 18.00 hours and 06.00 hours over three consecutive nights by trained collectors working in pairs using torches and aspirators. Four
pairs of collectors were rotated in 6-h shifts to reduce fatigue. Study houses were typical of regional adobe construction, with three partitioned rooms, tiled or palm-thatched roofs and earthen floors.

**ITN crossover study**

Four houses were enrolled into the crossover study. Two houses were provided with an ITN and the others with an untreated bednet. Treatments alternated each night in a repeat ABAB...AB design over 15 consecutive nights. One house dropped out of the study on day 8 (the owners travelled away), so an additional house from the seven houses surveyed (above) with similar sandfly abundance was enrolled from day 9. Sandflies were collected from 19.00 hours to 23.00 hours (the period of greatest sandfly activity), with 10-min hourly breaks, by trained health workers, one positioned inside the bednet, and one positioned ≤ 5 m away outside the bednet, using a torch and aspirator. Eight collectors were rotated between houses and collection positions (inside and outside nets). Sandfly captures were placed in cardboard cups and labelled according to the following capture location: alighting on the collector (hereafter referred to as ‘landing’); alighting on the net; alighting on the walls (outside nets), and immediate knockdown on the floor. The rate of 24-h mortality was measured by maintaining live caught sandflies on sucrose solution in cotton wool as the plug of the cups. To facilitate collection, nets were suspended over the floor of the bedroom rather than over the bed, and the floor under the net was lined with white plastic sheeting. Collectors searched for sandflies in all categories inside and outside the net throughout the collection period.

**Bednets**

The bednets were single-sized (180 × 220 × 120 cm), white polyester and pretreated with 25% deltamethrin EC (Smartnet; Commercial Marketing Strategies, Kampala, Uganda). Mesh size (64 per cm²) was large enough for sandflies to pass through. Control nets were similar but untreated.

**Sandfly identification**

Previous entomological surveys in the study area (Lainson et al., 1990; Kelly et al., 1997) demonstrated that the vast majority (> 93%) of CDC light trap sandfly catches in animal sheds (chickens and pigs), houses, dining huts and household yards are of *Lu. longipalpis* based on diagnostic characteristics of the spermatheca and pharyngeal armatures following Ryan (1986). These results were corroborated by sandfly identification of CDC and human landing catches in the crossover study houses during the same period (July–August) in the year previous to the current study (Barwa, 2002). A total of 98% of the 522 sandflies captured in that study were confirmed as *Lu. longipalpis*. Consequently, species identification was not attempted in 2003.

**Results**

**Crossover study**

A total of 1284 female sandflies were collected inside houses during the 30 trapping night crossover study (Table 1).

Adjusting for house and night effects, the mean absolute number of female sandflies captured per night in ITN houses

**Statistical analysis**

Female sandfly counts were highly overdispersed and could not be transformed to achieve normality or Poisson distribution; thus they were analysed by negative binomial regression using the log-link function. Proportions were analysed as binomial data using the logit-link function, and accounting for denominators. Reported estimates were adjusted for night and household unless otherwise stated. Crude Williams geometric mean sandfly counts were calculated as log(count) + 1, as shown in Table 1. Regression models were statistically tested using generalized linear modelling (GLM) and related routines in Stata Version 8.0 (Stata Corporation, College Station, TX, U.S.A.). In the crossover study, carry-over treatment effects of ITNs (A→B, B→A) between consecutive nights (Reed, 2004) were not expected, and sequence effects were removed by the multiple sequence crossover ABAB...AB design of the study. Period effects were accounted for by treating night as a covariate. Both house and night were forced into all analyses. Where nightly counts were too few to statistically compare using GLM, data were collapsed by treatment across nights and tested by chi-square test. Human ages were categorized into age classes of 0–5 years, 6–12 years and > 12 years. The 0–5-year-old group represents the age class most at risk of developing ZVL (Dye & Williams, 1993; Davies & Gavagni, 1999).

**Questionnaires**

Information on the use of hammocks, beds and bednets, children’s evening meals and bedtimes, household eating location and current pest control practices was obtained by interviewing the matriarch of 100 households in 10 villages. Census data were obtained on all residents of these households.

**Ethics**

Informed consent for the crossover study was obtained from all adults and, by proxy, children in study households. Impregnated bednets were given to all householders at the end of the study. The sandfly collectors were recruited from the endemic villages in the study area and were shown to be skin-test positive to *Le. infantum* (= *Le. chagasi*) antigen, which indicates a protective cellular response following previous exposure. Ethical clearance was obtained from the ethical committees of the London School of Hygiene and Tropical Medicine and the Instituto Evandro Chagas, in Brazil.

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was 14.4 (95% confidence interval [CI] 10.12–20.51), which was not dissimilar to the mean of 16.9 collected in houses with untreated nets (95% CI 11.93–23.94) (z = 0.62; NS, n = 60). The mean number collected per night under treated nets was 1.3 (95% CI 0.84–2.05), which was significantly fewer than the mean of 2.6 collected under untreated nets (95% CI 1.78–3.85) (z = 2.27; P < 0.023), and a similar percentage (96% (95% CI 7.10–12.92%) and 15.8% (95% CI 12.72–19.42%) of total nightly household captures, respectively (z = 2.74; P = 0.006). These data indicate that the insecticide increased the barrier effect of untreated nets by an average of 39.2% (95% CI 34.00–44.14%) ([15.8–9.6%]/15.8%). The insecticide also reduced the percentage of (collected) sandflies landing under nets from 71.4% (95% CI 59.91–80.87%) in untreated to 14.5% (95% CI 6.10–30.71%) in treated households, representing a reduction of 79.7% (61.93–89.90%) (z = 4.89; P < 0.001).

The reduced number of landing sandflies under ITNs was associated with a 4.9-fold (95% CI 2.06–11.43%) increase in the proportion of total sandflies under nets that alighted on the inside surface of treated nets compared with under untreated nets (z = 3.61; P < 0.001), in addition to a small increase in the crude proportion of sandflies knocked down (treated 7/44 vs. untreated 0/79; Fisher’s exact test: P < 0.001). The 24-hour mortality rate of all sandflies collected under ITNs was 97.7% (95% CI 92.52–98.93%), compared with 0% under untreated nets (treated 43/44 vs. untreated 0/79; Fisher’s exact test: P < 0.001). Mortality rates for the sandflies on the surface of the net and landing on collectors under ITNs were 32/32 and 4/5 compared with 0/23 and 0/56 under untreated nets, respectively (Fisher’s exact test in each case: P < 0.001).

The insecticide also affected the behaviour and survival of sandflies exterior to ITNs in the same room. The percentage of total collected sandflies that landed exterior to ITNs was reduced by 55.9% (95% CI 51.86–59.44%), from 48.6% (95% CI 43.82–53.50%) in untreated rooms to 21.5% (95% CI 17.88–25.85%) in ITN houses (z = 9.50; P < 0.001). The absolute number of sandflies landing exterior to ITNs was also reduced (2.4 [95% CI 1.42–4.20] vs. 6.9 [4.19–11.37]; z = 2.72; P < 0.007). This was despite a similar abundance of sandflies in rooms with the two respective treatments (untreated 14.0 [95% CI 9.58–20.38] vs. ITN 12.1 [95% CI 8.25–17.66]; z = 0.53; NS). The rate of 24-h mortality amongst sandflies collected exterior to ITNs was 67.7% (95% CI 62.00–73.00%), compared with 0.4% (95% CI 0.11–1.67%) of those collected exterior to untreated nets (z = 8.78, P < 0.001); all 351 sandflies recorded as landing in untreated houses survived > 24 h. No knockdown sandflies were collected outside treated or untreated nets.

The distribution of sandflies collected outside nets was also altered: the percentage of flies that were captured from the walls in ITN rooms (67.5% [95% CI 62.75–72.0%]) was approximately twice that recorded in untreated rooms (36.4% [95% CI 32.0–41.12%]) (z = 10.2, P < 0.001), whereas the fractions that alighted on the exterior surfaces of the nets were not dissimilar between treatments (ITN houses 10.8% [95% CI 8.19–14.36%] vs. untreated houses 14.0% [95% CI 11.17–17.70%]; z = 1.55, NS).

Within ITN houses, the 24-h lethality effect of the insecticide was greater inside than outside the net (z = 2.90; P = 0.004), and greater amongst flies alighting on the walls than landing outside the nets (z = 7.56, P < 0.001) (Table 2). The mortality rate amongst the 55 flies captured resting on the outside surface of the treated nets appeared even greater than that of flies captured resting on the walls (Yates corrected: 55/55 vs. 279/397; χ² = 20.6, P < 0.001).

**Indoor sandfly activity**

Indoor landing catches over three consecutive nights in two study houses revealed sandfly activity to commence soon after
Bedtimes, eating times and location

The majority (65.3%, 65/99) of families ate their evening meal inside the house. The remaining 34.7% (34/99) ate outside the house in the open or in the open-sided dining huts attached to the house. Dinner times ranged from 17.00 hours to 21.00 hours and peaked between 18.00 hours and 20.00 hours. Most families (95%, 94/99) ate before 20.00 hours (Fig. 2). Only 12% (9/75) of households reported that adults ate at different times (always 1 h later) to their children. According to children’s eating times, families eating outside tended to eat earlier (23/34 before 19.00 hours) than those eating inside (31/65) ($\chi^2 = 3.59, P = 0.058$), although they were demographically similar. Bedtimes amongst 0–5-year-old children ranged from 18.00 hours to 00.00 hours, and peaked between 20.00 hours and 22.00 hours: 12.3% (8/65) of children went to bed before 19.00 hours, and 4.6% (3/65) went to bed after 22.00 hours (Fig. 2).

Net use and net ownership

Members of 100 households were questioned about sleeping arrangements. Of the 528 household members, 384 (72.7%) slept in hammocks, 144 (27.3%) slept in beds. Of these, 12.2% (47/384) and 41.7% (60/144) used a hammock or bednet, respectively. Thus only 20.3% (107/528) of the population reported sleeping under a net, whereas the majority 63.8% (337/528) slept in a hammock without a net. Of the families questioned, 48% had at least one child aged 0–5 years; 30, 16 and two of the households contained one, two and three children in this age group, respectively. Stratified by age group, regular net use was reported for 33.3% (22/66) of 0–5-year-olds, 19.2% (33/172) of 6–15-year-olds, and 18% (52/289) of children aged ≥16 years ($\chi^2 = 8.01, P = 0.018$) (Fig. 3). None of the nets were reported to have been treated with insecticide at any time. All respondents regularly slept inside rather than outside the house.

A total of 34% of households owned at least one net, with a median of two (95% CI 2–4, range 1–8). Overall, 37% of households owned only hammocks, 9% owned only beds, and 54% owned one or more of both. Variations in ownership were 0–8 beds (median one), and 0–13 hammocks (median four) per household. In households that owned beds only, the mean number of residents was 3.7 (95% CI 2.25–5.23), which was marginally lower than the 5.3 (95% CI 4.40–6.26) in hammock-only households (unpaired t-test $t = 2.08, P = 0.052$), and lower than the 5.7 (95% CI 5.02–6.34) in mixed bed/hammock households (unpaired t-test $t = 2.79, P = 0.015$). The median ages and variance in median ages of these household groups were not dissimilar, suggesting that the average hammock-using household comprised a moderately larger family than in households using beds, and that this difference was not related to the age composition of the family.

Household insect control

About half (47%) of the 100 households reported using insect control measures other than nets: 46.8% (22/47) applied commercial insecticide (Baygon®, S.C. Johnson, Buenos Aires, B.A., Argentina, active ingredients: propoxur 0.75%, cyfluthrin 0.025%); 51.1% (24/47) generated smoke at the entrance to the

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**Table 2.** Adjusted 24-h mortality percentage rates for sandflies captured inside houses with insecticide-treated nets.

<table>
<thead>
<tr>
<th>Location</th>
<th>Inside net</th>
<th>n</th>
<th>Outside net</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td>97.7 (85.60–99.75)</td>
<td>44</td>
<td>63.8 (59.97–67.60)</td>
<td>597</td>
</tr>
<tr>
<td>Landing</td>
<td>32.4 (25.32–40.47)</td>
<td>145</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wall-resting</td>
<td>70.3 (65.61–74.65)</td>
<td>397</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

$n$, total number of sandflies in each category as described in Table 1; 95% confidence intervals in parentheses.

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The benefit of treated bednets to reduce indoor exposure to Lu. longipalpis was attributed in this study to increased sandfly 24-h mortality and excito-repellency, and improvement in the barrier effect of the net. Deltamethrin impregnation reduced sandfly entry into the net by approximately 39% compared with untreated nets, despite the mesh size being sufficiently wide to permit sandfly passage. The insecticidal activity also reduced the relative human-landing catch rate inside nets by approximately 80%, and was lethal to approximately 98% of sandflies. A significant increase in sandfly knockdown rate was also observed. Mortality was not observed under untreated nets; in this respect net use was no better than non-use of nets. However, there was an interesting trend towards lower landing rates inside compared with outside untreated nets (Table 1). The barrier effect of an untreated net relative to no net remains to be shown.

Insecticide-treated bednets also provided potential protection to non-net users in the same room by reducing both the relative and absolute landing rates to approximately half to a third of those in untreated rooms. This was associated with a doubling in the proportion of sandflies alighting on walls in ITN rooms, indicative of excito-repellency interfering with feeding behaviour. Lethality amongst these sandflies, and those collected in the room generally, was raised to 68–70% (Table 2). Improved protection against sandfly bites provided by impregnated nets or curtains compared with untreated materials is well documented (e.g. Alexander et al., 1995; Kroeger et al., 2002; Alexander & Maroli, 2003), whereas protection of non-net-users in a room containing an ITN has only been documented in one reported study of sandflies, in which there was a 42% relative reduction in human landing catches of Lu. youngi (an anthropophilic vector of cutaneous leishmaniasis in Colombia) (Alexander et al., 1995). More extensive experiments with Anopheles and Culex mosquito vectors, demonstrate that reduced feeding success is often due to insecticide-induced excito-repellency, which cause both fed and unfed mosquitoes to leave rooms containing ITNs (e.g. Lindsay et al., 1989; Soremekun et al., 2004), or to deterency effects of insecticide formulation volatiles that result in fewer mosquitoes entering ITN rooms or houses (Lindsay et al., 1991; Chandre et al., 2000).

In the current study, we found no evidence of either deterency or repellency. Firstly, similar numbers of sandflies were recorded per night in treated and untreated rooms, and alighting on the exterior surfaces of treated and untreated nets (Table 1). Secondly, although a significantly greater fraction of sandflies alighted on walls outside ITNs, these flies had a high mortality rate (see above) compared with the 0% rate in untreated houses, clearly indicating prior contact with the ITN. As expected, a similar or possibly higher rate of mortality was observed amongst sandflies collected directly from treated nets, despite the possibility that collectors reduced the insecticide contact time of these sandflies. The intensive collection effort, combined with the wider surface area to search in ITN rooms, probably explains the unexpected absence of knockdown sandflies outside treated nets. Thirdly, there was no evidence of increased landing rates outside treated nets compared with either inside treated nets or in untreated rooms. The study did not deploy exit traps to assess whether sandflies differentially escaped treated and untreated rooms/houses, nor was a no-net treatment group included to test if the barrier effect of the net (cf. insecticide) would divert bites to untreated rooms. The latter effect has been observed experimentally for Anopheles arabiensis Paton mosquitoes in Tanzania (Lines et al., 1987). Insecticide-treated net interventions inside houses are frequently associated with a shift towards increased mosquito exophagy and zoophagy, but not invariably so (reviewed by Takken, 2002). To our knowledge, there are no

![Fig. 2](image)

Marajó household dinner times (open bars) and 0–5-year-old children’s bedtimes (solid bars) in relation to activity of Lutzomyia longipalpis (both sexes), shown as cumulative geometric means (GM) of hourly human landing captures inside houses measured over 3 nights (dashed lines), and hourly Shannon trap catches in yards outside houses measured over 6 nights (solid lines). The x-axis shows the starting time of each 1-h interval. (Solid lines redrawn with permission from Quinnell & Dye, 1994b.)

![Fig. 3](image)

Regular net usage amongst members of 100 households stratified by age categories: 0–5 years, n = 66; 6–15 years, n = 172; > 15 years, n = 289.
published studies that show increased landing rates on non-net users in the proximity of ITNs. Negative displacement effects of pyrethroids are more often associated with their application to substrates other than bednets (albeit tested rarely). Previous studies in our study villages in Marajó demonstrated that blanket coverage of chicken sheds with lambdacyhalothrin ME (20 mg a.i. m\(^{-2}\)) resulted in increased CDC light trap catches in nearby unsprayed open dining huts compared with controls, suggesting diversion onto unprotected humans (Kelly et al., 1997).

In general, ITNs are expected to be most successful at reducing intradomiciliary transmission when the vector is active after people are in bed under a net, and when the vector is predominantly endophagic. Conversely, ITNs may not be as effective when the indoor : outdoor ratio of the vector population is low. In Marajó, *Lu. longipalpis* shows peak indoor activity at 19.00–23.00 hours (Fig. 2), which is similar to the time of initial sandfly activity inside animal sheds (Quinnell & Dye, 1994b). Indoor activity appears to lag by approximately 1–2 h behind outdoor activity, which starts at 17.00 hours and peaks at 18.00–20.00 hours (Shannon trap catches [Quinnell & Dye, 1994b] shown in Fig. 2). *Lu. longipalpis* is estimated to be nine times more abundant in animal sheds (principally chicken sheds) than inside houses after adjusting for trapping efficiency (Quinnell & Dye, 1994b), which is partially explained by their tendency not to enter well constructed houses (Quinnell & Dye, 1994a), and their lack of strong innate host preference for humans (Quinnell et al., 1992). Other members of the *Lu. longipalpis* complex show similar opportunistic feeding behaviour. Immunodiagnostic evidence from Marajó suggests that transmission probably occurs predominantly outside the house: seroprevalence and infection incidence in our study villages in Marajó demonstrated that blanket coverage of chicken sheds with lambdacyhalothrin ME (20 mg a.i. m\(^{-2}\)) resulted in increased CDC light trap catches in nearby unsprayed open dining huts compared with controls, suggesting diversion onto unprotected humans (Kelly et al., 1997).

In conclusion, ITNs show promising entomological effects in ZVL transmission, persuading this population to use nets – and to impregnate them if longlasting insecticidal nets (LLINs) are not the first choice – would require a well structured co-ordinated education programme at the primary health care level, coupled with free provision of nets or social marketing. In other leishmania-endemic regions of Amazon Brazil, 96% of the population sleep under nets, and only 0.7% sleep in hammocks (Alexander et al., 2005). A common limitation of community-based ITN programmes is the failure to re-impregnate; thus untreated nets might be more appropriate provided they are maintained in good condition (Lindsay et al., 1989). Clearly, this would avoid issues of re-impregnation, potential selection for insecticide resistance, and associated costs, although at a loss of efficacy as suggested by this study. The recent appearance of a ‘dip-it-yourself’ insecticide formulation for longlasting treatment of nets (Yates et al., 2005) may help to alleviate some of these constraints.

In conclusion, ITNs show promising entomological effects for the reduction of indoor ZVL vector human-landing rates in this endemic setting, although they are unlikely to have any impact on outdoor sandfly densities. Alternative pyrethroid insecticide applications that protect the domestic dog (Xiong et al., 1994, 1995; Gavagni et al., 2002b) may be more effective than ITNs in reducing transmission as dogs are the important zoonotic reservoir of *Le. infantum*. Given the operational difficulties with the current insecticidal spraying programme, ITNs might be considered an alternative or complimentary intervention.

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