Raised houses reduce mosquito bites

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Abstract

Background: In many parts of continental Africa house construction does not appear to impede entry of malaria vectors and, given their generally late biting cycle, the great majority of transmission takes place indoors. In contrast, many houses in São Tomé, 140 km off the coast of Gabon, are raised on stilts and built of wooden planks. Building on stilts is a time-honoured, but largely untested, way of avoiding mosquito bites. Exposure may also be affected by mosquito activity times and age composition of host-seeking females. A study was therefore undertaken on the island of São Tomé to determine if exposure to Anopheles gambiae, the only vector on the island, varied with house construction or time of the night.

Methods: A series of all-night landing collections were undertaken out of doors at ground level, inside houses at ground level, on the verandas of, and inside houses built on stilts. The gonotrophic age of an unselected sample of insects from the first three hours of landing collection (18:00–21:00) was determined by dissection. In addition, 1,149 miniature light-trap collections were obtained from 125 houses in the study area. Numbers collected were related to house construction.

Results: Biting of An. gambiae took place primarily outside at ground level. Less than one third of biting occurred inside houses. Houses built on stilts had half the number of An. gambiae in them compared to those built at ground level. Conversely houses with an eaves gap had more An. gambiae in them than houses without such a gap. Gonotrophic age did not affect house entry rates in An. gambiae. House construction affected Culex quinquefasciatus less than An. gambiae. Mean density per house, derived from a series of 1,490 randomly assigned light-trap collections, was over-dispersed with 18% of houses having 70% of the vectors.

Conclusion: House construction plays an important role in determining exposure to malaria vectors in São Tomé. Neighbours can have very different exposure levels. Recommendations for improvement in control are given.

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Background
In most areas of Africa the great bulk of feeding by the principal malaria vectors, *Anopheles funestus* and *Anopheles gambiae*, takes place in the latter part of the night, indoors [1]. Entry rates and hence transmission are affected by house construction. Thus, houses built of brick and with zinc roofs are associated with lower levels of malaria-associated anaemia in Tanzania compared to poorly built mud walled houses [2]. Similarly, in Sri Lanka house construction appears to be an important determinant of exposure to malaria [3]. Improvements in house construction, in particular screening against mosquito entry, have, therefore, long been recommended as a way of reducing exposure [4].

One way to do this is to sleep on a raised structure or in a house built on stilts [4]. The practise was described by Herodotus in Egypt 2450-2420 B.P. [5] and has been independently adopted by a number of different groups of people. For example, in Northern Vietnam the hill people build their houses on stilts to avoid *Anopheles minimus*, which seldom fly more than 2–3 m above the ground [6]. The anophelines feed on the animals kept under the houses and avoid the people. When people from the low-lands move into the hills they bring their culture with them. Their houses are built on the ground and animals are stabled alongside. As a result, *An. minimus*, which will feed on cattle but prefers humans, becomes an active vector of malaria [6].

Malaria was introduced in the archipelago of São Tomé and Príncipe shortly after colonization in the early 1500’s [7]. Sporozoite rates in *An. gambiae*, the only vector present on the islands, are generally less than 1% [8,9] and, despite high vector densities, the disease reaches only meso-hyperendemic levels [10,11]. A reason for this may be the propensity for the vector to feed on non-human hosts, particularly dogs and pigs [12]. These feeds are all taken out of doors. Given that *An. gambiae* is generally thought to prefer human blood and is considered to be ‘indifferent’ to the location of the host [1], the observed blood feeding patterns suggest that entry into houses in São Tomé for the mosquito is difficult. According to an early visitor, many of the free African inhabitants on the islands built their houses of sawn timber planks on stilts, to avoid ground-level humidity and mosquito bites [7]. This type of construction still predominates on both islands. However, in a recent survey, people living in houses raised on stilts in Príncipe, the smaller of the two islands that make up the two main islands in the archipelago, did not have a lower prevalence of malaria than people living at ground level [13].

Present vector control measures are all based against mosquitoes biting indoors. Should a high proportion of infections be acquired outdoors, alternative strategies of vector control need to be considered. In this study we examine the influence of house construction on biting rates of the local *An. gambiae*. The gonotrophic age of insects was also determined in order to assess whether the protective effect, if any, of building a house on stilts is reduced because older insects bite at higher levels [14]. Results are discussed with respect to novel vector control strategies that might be adopted on the islands.

Methods
Studies were undertaken in Riboque, a peri-urban area close to the capital city centre. Riboque has numerous houses with sown timber walls and zinc-roofed houses; many built on stilts of varying heights, in close proximity to each other. Banana plants and a variety of fruit trees grow throughout the area and a small stream (Lukumi) runs through the middle. During the rainy season the stream is liable to flooding and there are extensive swamp areas. Climate is equatorial with annual mean temperatures around 25°C and relative humidity of 80%. There are two short dry seasons, June-August and December–January.

A series of all-night landing collections were undertaken out of doors at ground level, inside houses at ground level, on the verandas of, and inside houses built on stilts in the wet season of 1997. Mosquitoes were collected with an aspirator as they landed on the exposed lower legs of adult men working in six-hour shifts, all of who were offered prophylaxis according to the protocols of the Centro Nacional de Endemias of São Tomé (CNE). Following the initial all-night collections a series two-hour landing collections (from 2100–2300 h) were undertaken each month outside, at a ground level sentinel site, and numbers were compared to those caught in sentinel light-traps.

Each hour’s collection was kept in individual containers and the gonotrophic age of an unselected sample of insects from the first three hours of landing collection (18:00–21:00) was determined by dissection of the female’s ovaries and spermathecae following the schema described by Charlwood et al. [8].

Host-seeking mosquitoes were also caught with Centre for Disease Control (CDC) miniature light-traps run all night close to the bed of sleepers protected by an untreated mosquito net [8]. In 1997 and 1998, houses for light-trap collections were selected at random, from a list of 25 houses. Five traps were run per night. An additional trap was run in a sentinel house. From May 1997 the number of houses in the study increased to 157. New sentinel houses were employed in the event that houses in use became unavailable.
The data from the all-night landing collections were balanced by eliminating measurements outside the timeframe over which indoor catches in elevated houses were obtained (18 March – 8 April 1997) and also houses oversampled during this timeframe. In all 93 house-hours were systematically selected for each classification (inside/ outside and ground level/elevated). Mosquito counts were log-transformed and a linear model, using S-PLUS 4.5 [15], was fitted to the log-transformed counts for An. gambiae and Cx. quinquefasciatus.

**Results**

Sixteen all night landing collections were performed out of doors at ground level, 22 inside houses at ground level, 15 on the verandas of, and eight inside houses built on stilts. A total of 8,926 An. gambiae, 6,539 Culex quinquefasciatus, 15 Aedes aegypti and a small number of unidentified culicines were collected. Differences in mosquito density were observed according to location of collection site and time of collection (Fig 1). More An. gambiae were collected outdoor biting to indoor biting and biting at ground level to biting at elevated levels. Table 1 shows linear model results for each species.

Gonotrophic age distribution of the 2, 818 An. gambiae dissected from the first three hours of collection was similar among different collection sites (Table 2).

From March 1997 to October 1999, 1,149 light-trap collections were obtained from 125 bedrooms in the study area. The number of An. gambiae and Cx. quinquefasciatus in light-traps were not significantly correlated (Spearman rank correlation coefficient on log-transformed ($x+1$) numbers, rho = 0.166, n = 125, p = 0.065). The distribution of An. gambiae was over-dispersed with 70% of the mosquitoes being collected from 18% of the houses (Fig 2a). Densities varied greatly between neighbouring houses according to construction. For example, eighteen-fold differences in mean densities of An. gambiae per night were recorded between houses five meters apart one of which was built at ground level whilst the other was built on stilts two metres high (mean per night = 68.7, s. d. =

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**Table 1:** Mean numbers of *Anopheles gambiae* and *Culex quinquefasciatus* collected per hour in landing catches, according to location of collection site, Riboque, São Tomé.

| Mean number collected per man-hour (95% CI) | Outdoor ground level | Veranda | Indoor ground level | Indoor upper level |
|--------------------------------------------|----------------------|---------|---------------------|-------------------|
| An. gambiae                                | 16.51 (12.8–21.2)    | 8.10 (5.5–11.7) | 3.58 (2.9–4.4)   | 2.38 (1.7–3.3)   |
| Cx. quinquefasciatus                       | 5.11 (3.9–6.6)       | 5.14 (3.5–7.4) | 6.18 (5.1–7.4)   | 4.23 (3.2–5.5)   |

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**Table 2:** Physiological age of *Anopheles gambiae* collected in landing collections, according to location of collection site, Riboque, São Tomé.

| Physiological age (An. gambiae) | Outdoor ground level | Veranda | Indoor ground level | Indoor upper level | $\chi^2$ |
|--------------------------------|----------------------|---------|---------------------|-------------------|---------|
| Virgin                         | 167                  | 91      | 49                  | 14                | 0.30    | n.s.   |
| Plug positive                  | 103                  | 44      | 26                  | 3                 | 0.84    | n.s.   |
| Nulliparous II                 | 38                   | 21      | 12                  | 3                 | 0.57    | n.s.   |
| Parous with Sac                | 193                  | 100     | 58                  | 6                 | 0.04    | n.s.   |
| Parous No-sac                  | 90                   | 55      | 39                  | 8                 | 0.39    | n.s.   |
| Proportion parous              | 0.48                 | 0.50    | 0.52                | 0.41              |         |        |
The concentration in numbers of *Cx. quinquefasciatus* was not so apparent (fig 2b). The number of both *An. gambiae* and *Cx. quinquefasciatus* in light-traps were negatively correlated with the house being built on stilts (Spearman rank correlation coefficient on log transformed numbers (x+1), rho = -0.71, n = 25, p = <0.0001 and rho = -0.54, n = 25, p = 0.006 respectively). The number of *An. gambiae* in light-traps, but not the number of *Cx. quinquefasciatus*, was positively associated with the presence of an eave gap (respective Spearman rank correlations rho = 0.567, n = 25, p = 0.0032 compared to rho = 0.177, n = 25, p = 0.4). There was, however, no association for either species between the number of windows, doors or inhabitants that a house had and the number of mosquitoes caught (data not shown).

A mean of 16.2 (s.d. 5.8) light-trap collections were run in sentinel houses per month and a mean of 7 (s.d. 14) two hour landing collections undertaken per month. The relationship between mean numbers of *An. gambiae* caught per month in sentinel collections and per hour in routine outdoor landing catches is shown in Fig 3. Mean monthly number of mosquitoes caught by both methods were highly correlated (y = 0.196x + 15.8, R2 = 0.776).

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Discussion
The ancient Egyptians were right. People in houses raised on stilts suffer fewer mosquito attacks than people living on the ground. House construction affects indoor exposure to malaria vectors in suburban São Tomé just as it does in rural Vietnam [6]. Most An. gambiae were caught outside, at ground level and the least were caught inside houses built on stilts. Houses built at ground level had more mosquitoes in them than houses built on stilts. This can give rise to an 18-fold difference in exposure to malaria vectors between houses less than 10 m apart. Not unexpectedly higher mosquito numbers in light-traps were associated with houses in which there was an eaves gap [16]. In Príncipe, people living in houses with an eaves gap were also associated with a higher risk of infection with malaria parasites than those from other houses [14]. House construction or location of the host (inside or outside) affected Cx. quinquefasciatus less than it did An. gambiae. While Cx. quinquefasciatus did show a weak preference for ground level biting to elevated levels there was no preference for indoor over outdoor biting and the poor overall fit suggested that other factors, not accounted for in the analysis, were important. At the time of the study biting rates for Cx. quinquefasciatus were, on the whole, lower than for An. gambiae, which could have been the result of the peak season for Cx. quinquefasciatus occurring at a different time of the year. The wide variation of biting rates for An. gambiae among individual houses suggests that even in this species more parameters could be added to the model.

Light-trap collections undertaken inside houses and outdoor landing catches undertaken in high density areas were well correlated. Light trap collections may therefore be a suitable alternative to landing collections in estimating seasonal/longitudinal fluctuations of An. gambiae, thus avoiding unnecessary exposure of collectors while performing landing captures. The age structure, at least in the first three hours of the night, of An. gambiae caught inside or outside or in ground-level or elevated locations were similar. Virgin female An. gambiae, which have to fly at least two meters above the ground in order to mate [17], appear to seek hosts by flying close to the ground in much the same way as other age groups. Given that the age structure of the insects at ground level outside was similar throughout the night [18] it is likely that the age structure of insects caught in the other locations also remains constant throughout the night. The constancy of parous rates implies that potentially infective insects will bite at all times of the night, regardless of location.

At the time of the study, many people in São Tomé watched communal television out of doors. Similarly, potential purchasers of the catch will wait on the beach for the return of the fishermen at night and, depending on the tides, can spend many hours outside. The availability of cheap repellents [19] would reduce risk in such situations.

The concentration of mosquitoes in a small number of houses also implies that, if high risk houses can be easily identified and if it is socially acceptable, some sort of focal control might be feasible. Spraying high density houses (with an insecticide differing from the one used on nets) would expose much of the house-entering fraction of the mosquito population to two different insecticide groups and reduce the likelihood of resistance developing, much as combined therapy might reduce resistance developing in parasites.

Spraying does not, however, provide personal protection and cannot be easily implemented in a house-by-house way. Bednets have been widely available in São Tomé since 1995. Poorer households are still more likely to be inadequately protected and at low coverage rates unimpregnated bednets may even increase transmission in children [20]. Covering the eaves of houses with insecticide impregnated netting or installing netting ceilings has been shown to reduce mosquito entry rates and malaria transmission in continental Africa [21,22]. Such systems could be installed in houses with eaves in São Tomé. This would also provide a measure of protection to those who cannot afford bed nets.

Abbreviations
CDC – Centres for Disease Control, CI – Confidence Interval CNE – Centro Nacional de Endemias, s. d. – standard deviation

Authors’ contributions
Charlwood – Designed the study, supervised collections, dissected the mosquitoes and wrote the manuscript
Pinto – Dissected the mosquitoes and wrote the manuscript
Ferrara-Undertook the statistical analyses
Sousa – Dissected the mosquitoes and wrote the manuscript
Ferreira – Provided logistical support in São Tomé
Gil – Provided logistical support in São Tomé
Rosario – Provided logistical support in Portugal

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