How reliable are light traps in estimating biting rates of adult *Anopheles gambiae* s.l. (Diptera: Culicidae) in the presence of treated bed nets?

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Abstract

The sampling efficiency of light trap catches relative to human bait catches in estimating biting rates of the mosquito *Anopheles gambiae* Giles was investigated in two types of community in southern Sierra Leone: (i) where most of the inhabitants slept under treated bed nets; and (ii) where most of the inhabitants slept without bed nets. The number of female *A. gambiae* mosquitoes caught in these communities by light trap was strongly correlated ($r \geqslant 0.72$) with those from corresponding human biting catches performed either on the same or adjacent nights. It was found that the relative sampling efficiency of light traps varied slightly but significantly with mosquito abundance in villages with treated bed nets, but not in those without them. Nevertheless, the relationship between relative sampling efficiency and mosquito abundance did not differ significantly between the two types of village. Overall, there was insufficient evidence to show that the presence of treated nets altered the relative efficiency of light traps and any bias was only slight, and unlikely to be of any practical importance. Hence, it was concluded that light traps can be used as a surrogate for human bait catches in estimating biting rates of *A. gambiae* mosquitoes in the two communities.

Introduction

The human biting rate of mosquitoes is an essential component of vectorial capacity and entomological inoculation rates, the two most important concepts for describing and comparing transmission intensities in entomological terms (Burkot & Graves, 1995). Measuring the

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biting rates of mosquitoes, therefore, constitutes a very important aspect of entomological monitoring of vector control interventions, such as insecticide-treated nets.

The most direct way of estimating biting rates is by human biting catches (HBC), because with this method mosquitoes are caught while engaged in the very act of biting (Service, 1993). However, this method has logistical problems. For example, it is difficult to supervise, expensive, labour intensive and requires skilful catchers. It also raises an ethical problem, because it may expose the catchers to more mosquito bites and hence an increased risk of contracting malaria. These objections have led to a search for surrogate methods, such as light trap catches (LTC), that

have the potential to provide indirect but reliable estimates of human biting rates (Lines *et al.*, 1991).

Odetoyinbo (1969) made the first comprehensive study of light-traps as a sampling method, and found that they could be used for assessing night-time densities of different mosquito species, but added that they were not reliable for assessing human biting rates. This method was later modified by Garrett-Jones & Magayuka (1975). They tried placing the light trap beside an occupied untreated bed net and found that its efficiency for assessing human biting rate was greatly improved. They also found that most of the mosquitoes caught were unfed females, suggesting that this might be a suitable method of sampling blood-seeking mosquitoes. Since then, various studies have investigated the reliability of light-traps for measuring human biting rates of various mosquito species, including *Anopheles fluviatilis* James (Gunasekaran *et al.*, 1994*), Anopheles albitarsis* Lynch Arrobálzoga, *Anopheles triannulatus* Neiva & Pinto, *Anopheles neomaculipalpus* Curry (Rubio-Palis & Curtis, 1992) and *Anopheles gambiae* Giles (Diptera: Culicidae) (Lines *et al.*, 1991; Faye *et al.*, 1992; Mbogo *et al.*, 1993; Davis *et al.*, 1995). Most of the studies carried out in Africa showed that a light trap hung beside an occupied untreated bed net is an efficient and unbiased way of estimating the human biting rate of *A. gambiae* populations (Lines *et al.*, 1991; Davis *et al.*, 1995; Costantini *et al.*, 1998).

In Kenya, Mbogo *et al.* (1993) reported that light traps did not provide an adequate estimate of the human biting rate of *A. gambiae* mosquitoes. In this case, the absence of a clear relationship between the two catching methods was attributed to the relatively low mosquito density in this study (Smith, 1995). In other cases, however, light traps catches have failed to show a clear correlation with human bait catches despite the presence of high or moderate mosquito densities and, in such cases, light traps catches are clearly not suitable as substitutes for human bait catches (e.g. Rubio-Palis & Curtis, 1992)

In locations where good relationships have been found between light trap and human bait catches, light traps have been used in the evaluation of bed net trials to estimate changes in mosquito biting rates (e.g. Curtis *et al.*, 1998; Maxwell *et al.*, 1999). So far, however, it has not been shown whether light traps can provide a reliable estimate of biting rates in communities where most people sleep under treated bed nets. In these studies, as in this one, the nets used by people in light trap sampling rooms remained untreated even when everyone else in the village was using treated nets. Nevertheless, it is known that insecticides on the nets can repel mosquitoes (Lindsay *et al.*, 1992; Lines, 1996) and might cause mosquitoes to be driven away from the sampling house resulting in reduced light trap catches and thus may under-estimate true biting rates.

This paper therefore investigates the reliability of biting rate estimates of *A. gambiae* mosquitoes obtained from light trap catches in both villages with treated nets and those without, by comparing them with those obtained from matched human bait catches.

Materials and methods

Study area

The study was undertaken in 16 villages in the north-eastern part of Bo District, southern Sierra Leone. These villages were involved in a bed net trial investigating the effect of lambdacyhalothrin treated bed nets on malaria morbidity in children, and on malaria transmission. These villages and the trial itself have been described elsewhere (Peterson *et al.*, 1993; Magbity *et al.*, 1997; Marbiah *et al.*, 1998). Bed nets treated with 10 mg a.i. m^{-2} of lambdacy-halothrin were supplied to all the sleeping places in eight of these villages, while the other eight remained as controls without bed nets.

Chromosomal studies by Bockarie *et al.* (1993) revealed that the *Anopheles gambiae* s.l. mosquitoes in the area were the forest form of *Anopheles gambiae* s.s.

Mosquito sampling methods

In each village, mosquitoes were sampled using human bait catches once a month on the veranda of a designated house by a team of four mosquito catchers working in pairs, on alternate 3-h shifts. Mosquito collection started from 1900 h and continued till 0700 h the next morning.

Corresponding light trap catches, using CDC 512 Miniature light traps and batteries (6V, 10A) were carried out on either the same night, or an adjacent night to the human bait catches. The light trap catches were carried out beside occupied untreated bed nets in three other designated houses as described by Lines *et al.* (1991). In villages with insecticide treated nets, the nets in the same rooms as the light trap were replaced with untreated nets, but all other sleeping places retained their treated nets. In each sampling room, a single light trap was suspended about 1.5 m from the floor and about 0.2 to 0.5 m from the bed net. The traps were turned on at about 1900 h and off at about 0700 h the following morning by a project staff member, who enquired whether the sleepers noticed any malfunctioning of the trap during the night. Catches from traps that malfunctioned were discarded.

Statistical analysis

The total number of *A. gambiae* mosquitoes caught by three light traps was compared with the number caught by two human baits in matched human bait catches on either the same or an adjacent night, by regression analysis, using STATA statistical software (StataCorp, 1995). The analysis was performed for two different sets of data: one comprised cases with catch $\geqslant 0$ and the other included only cases when both catching methods caught at least one mosquito. Data (light trap and corresponding human bait catch) for nights when either one or more light traps malfunctioned were excluded from the analysis.

The daily number of mosquitoes (x) caught by each sampling method was transformed to $\log_{10}(x + 1)$, to normalize the distribution. The relative sampling efficiency was measured as the ratio of the number of mosquitoes caught by the light trap (LTC) to the number caught by the human baits (HBC) (Altman & Bland, 1983).

To test whether the relative sampling efficiency of light traps was affected by mosquito density, the relative sampling efficiency, calculated as *log(LTC + 1) − log(HBC + 1)*, was plotted against a joint estimate of mosquito abundance, calculated as *(log(LTC + 1) + log(HBC + 1))/2* (Altman & Bland, 1983). In this method, a significant regression coefficient implies that the relative sampling efficiency of light traps tends to increase or decrease at higher mosquito

abundance, i.e. it is biased as a means of measuring changes in density.

Analysis of variance (ANOVA) was performed to determine the effect of treatment on the relative sampling efficiency of light traps.

Results

Mosquito abundance

A total of 2299 female *A. gambiae* mosquitoes were caught by both sampling methods. About 45% of all the mosquito sampling occasions yielded no mosquitoes and less than 20% yielded more than five mosquitoes (fig. 1).

Relationship between the number of mosquitoes caught by light traps and human baits

The relationship between biting rates (including zero catches) estimated from light traps and human bait catches in both villages with and without treated nets is shown in fig. 2. The Pearson correlation coefficient (r) for villages with treated nets was 0.751 ($P < 0.001$) and 0.722 ($P = 0.0001$) for those without them. When all cases with zero catches were excluded, the corresponding Pearson correlation coefficients were 0.724 ($P < 0.001$) for villages with nets and 0.671 ($P = 0.001$) for those without. These clearly indicated a strong positive relationship between light traps and human bait catches in both villages with treated nets and those without. Moreover, when the data set included cases with zero catches, the slope of the regression line for villages with treated nets and those without were 0.88 (95% C.I. = 0.72 to 1.05) and 0.78 (95% C.I. = 0.60 to 1.01) respectively, not significantly different from each other. This implies that

three light traps caught almost as many mosquitoes as matched human bait catches, in both villages with treated bed nets and those without. When the analysis was repeated excluding cases with zero catches, the corresponding regression slopes for villages with treated nets and those without were 0.59 (95% C.I. = 0.44 to 0.73) and 0.57 (95% C.I. = 0.35 to 0.79) respectively, also not significantly different from each other.

The geometric mean ratio of matched light trap and human bait catches (including zero catches) in villages with treated nets and those without were 0.90 (95% C.I. = 0.77 to 1.06) and 1.06 (95% C.I. = 0.88 to 1.26) respectively. Both ratios were not significantly different from unity, confirming the result from the regression analysis, that on average, three light traps caught approximately as many mosquitoes as two human baits in villages with and without treated nets.

The effect of treated bed nets on the relative sampling efficiency of light traps was assessed by analysis of variance performed on the log-transformed ratios of the paired mosquito catches, including cases with zero catches (table 1). The results confirmed that there was no significant difference in mean log-ratios between villages with treated nets and those without. There was, however, a slight indication of a difference in mean log-ratios between individual villages, and a considerable proportion of the variance between log-transformed ratios was explained by inter-village variation. This result was not affected by the exclusion of cases with zero catches from the analysis.

A possible explanation for the differences in sampling efficiency between villages could be that the relative sampling efficiency of the light trap was dependent on the mean mosquito abundance, and that villages differed in mosquito abundance. To test for the effect of mosquito abundance on sampling efficiency, the relative sampling efficiencies of the
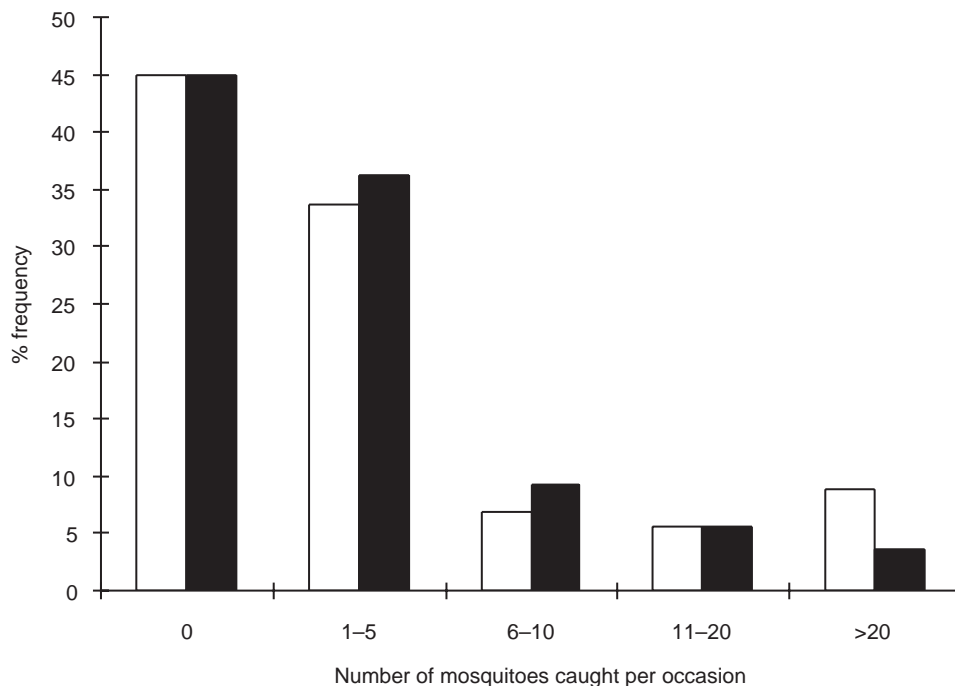

Fig. 1. Frequency of occasions which various numbers of *Anopheles gambiae* mosquitoes were caught by each sampling method (□, human bait; ■, light trap).
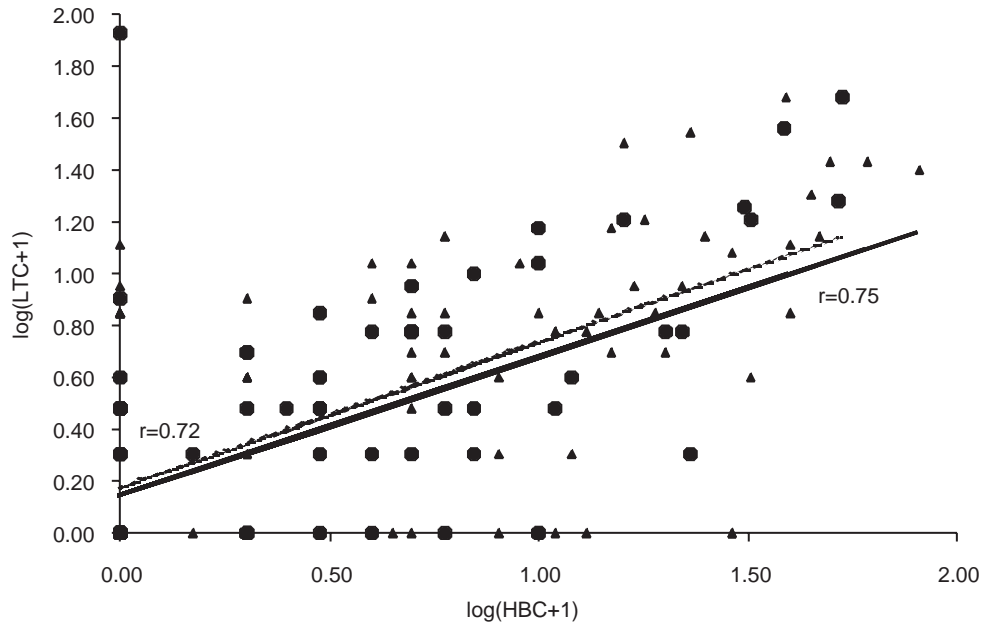
Fig. 2. The relationship between human biting catches (HBC) and matched light trap catches (LTC) of *Anopheles gambiae* mosquitoes in villages with (▲) and without (●) treated bed nets. The lines show the fitted regressions in villages with (—) and without (---), respectively.

Table 1. ANOVA on the log-transformed ratios between light-trap catches (LTC) and human bait catches (HBC) of *Anopheles gambiae*, calculated as (*log(LTC + 1) − log(HBC + 1)*), in villages with and without treated bed nets.

Source	Partial SS	d.f.	MS	F	P
Between treated and untreated village groups	0.807	1	0.807	4.93	0.0275
Between villages within treatment groups	5.349	24	0.223	1.30	0.128
Within villages	36.196	221	0.163		
Total	41.745	246	0.170		

light trap were plotted against mosquito abundances, including cases with zero catches (fig. 3). In villages without treated nets, the regression slope was not significantly different from zero (b = −0.08; 95% C.I. = −0.29 to 0.12;), indicating that the relative sampling efficiency of the two methods did not tend to vary over the observed range of population densities. However, in villages with treated nets, the regression slope (b) was −0.19 (95% C.I. = −0.358 to −0.072), slightly less than zero. When the analysis was repeated excluding cases with zero catches, the corresponding results were similar. The regression slope for villages without treated nets was not significantly different from zero (b = −0.20; 95% C.I. = −0.501 to 0.107), while that for villages with treated nets was slightly significantly different from zero (b = −0.236; 95% C.I. = −0.438 to −0.033). This indicated a slight tendency for the sampling efficiency of light traps to decline relative to human bait catches at high mosquito densities. Nevertheless, the upper confidence limits in treated villages were very close to zero, and the slopes of the regression lines for villages with nets and those without nets did not differ significantly from each other.

Discussion

This study showed that the number of adult female *A. gambiae* mosquitoes caught in light traps was strongly positively correlated with those obtained from human bait

catches performed on either the same or an adjacent night, in both villages with treated nets and those without. There was no evidence that the relative sampling efficiency of light traps varied with mosquito abundance in villages without treated nets. The results therefore confirm those of Lines *et al*. (1991) in Tanzania and Costantini *et al*. (1998) in Burkina Faso, that light traps can be used as an unbiased estimator of biting density in villages without treated bed nets. In villages with treated nets, there was some evidence that the presence of treated nets may marginally reduce the relative sampling efficiency of light traps at high population densities. However, it appears that the magnitude of the effect of treated nets on biting rates obtained from light traps was not large enough to be of practical significance, at least at the moderate population densities observed in this study.

In this study, the light trap catches were carried out in houses that in most cases contained treated nets, although the nets in the same rooms as the trap were always untreated. It is possible that if light trapping was carried out in houses without treated nets in all the rooms, the impact of the presence of the treated nets in nearby houses would have been completely insignificant (Lindsay *et al.*, 1992).

The estimated geometric mean ratio of light trap catches and human bait catches in both villages with treated nets and those without were close to unity, signifying that the total number of *A. gambiae* mosquitoes caught by three light
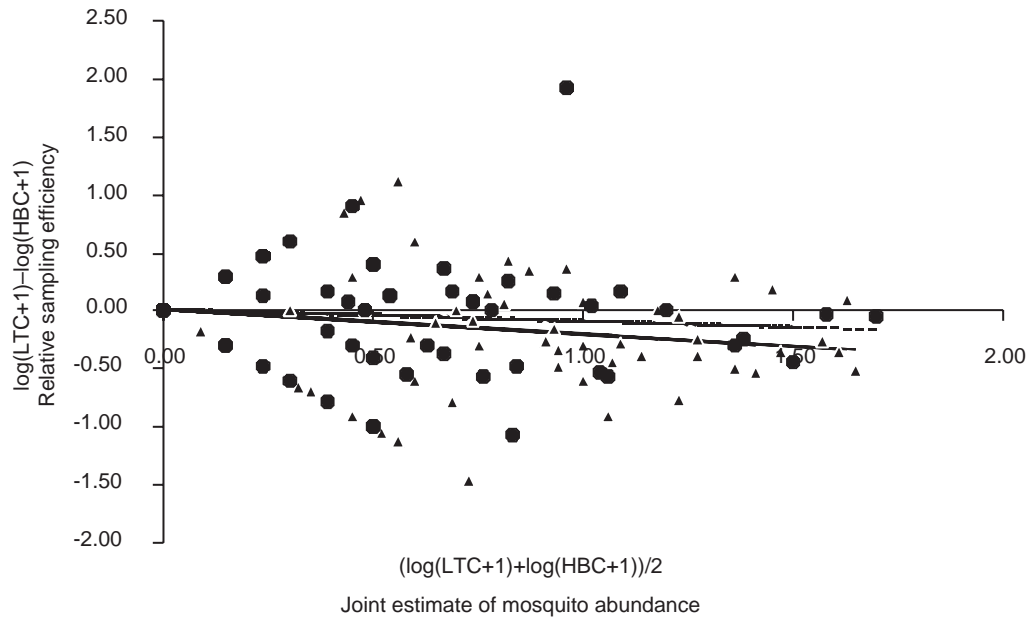
Fig. 3. The relationship between the relative sampling efficiency of light trap catches (LTC) ($log(LTC + 1) - log(HBC + 1)$) and mosquito abundance $((log(LTC + 1) + log(HBC + 1))/2)$ in villages with (▲) and without (●) treated bed nets. The lines show the fitted regressions in villages with (—) and without (– – –), respectively.

traps operating in different bedrooms for the whole night was not significantly different from that caught by two human baits working from dusk to dawn on the same or an adjacent night. This result is consistent with that obtained for *A. gambiae* by Lines *et al.* (1991) in nearby villages in northern Tanzania. In Burkina Faso, Costantini *et al.* (1998) found that the number of mosquitoes caught by one light trap was equivalent to that caught by one human bait.

The conclusion from this study is that biting rates from three light traps can replace those obtained from human bait catches performed on either the same or adjacent night, both in villages with and without treated bed nets.

In this study, about 45% of the sampling occasions yielded no mosquitoes. Smith (1995) claimed that in such cases the transformation log(x + 1) may not approximate to log (x), and had suggested the use of a Poisson regression. However, he did not elaborate on how this could be carried out, and moreover, Poisson regression could not be applied to the method of Altman & Bland (1983). Therefore to confirm the results obtained in this paper, the method of Altman & Bland (1983) was applied when zeroes were included or excluded from the data. The results obtained from these two different sets of data were very similar. In a recent paper, Hii *et al*. (2000) used a novel statistical approach to investigate the relative efficiency of light traps for estimating the abundance of some anophelines in Papua New Guinea. By assuming a Poisson distribution and using the statistical package BUG, they showed that light traps could replace human bait catches for some mosquito species but not for others. However, it is not clear whether their statistical method is superior to that of Altman & Bland (1983) and, until a reliable method is developed, most people are likely to continue using the transformation, log(x+1), even for very low mosquito counts. The need for a robust statistical technique for method comparison is critical. There is, therefore, an urgent need for more research in this area.

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