A COMPARISON OF TWO COLLECTION METHODS FOR ESTIMATING ABUNDANCE AND PARITY OF *ANOPHELES ALBIMANUS* IN BREEDING SITES AND VILLAGES OF SOUTHERN MEXICO

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ABSTRACT. The abundance and age structure of Anopheles albimanus populations were estimated by UV updraft light traps and human landing catches within villages and in nearby breeding sites of southern México. Four villages and 5 breeding sites were selected for the study. Light trap and human landing catches were simultaneously carried out in each breeding site and each village. Anopheles albimanus was the most abundant malaria vector caught in breeding sites and in villages. Significant differences in overall An. albimanus abundance among villages and among breeding sites were detected only by human landing catches. In both villages and breeding sites, more mosquitoes were captured by 1 human bait (34.3 \pm 6.3 and 14.6 \pm 2.9, respectively) than by one light trap (15.9 \pm 3.3 and 2.4 \pm 0.3 respectively) collection. After pooling, no significant differences were detected in the abundance estimated by each method in breeding sites and villages. A significant correlation of numbers of specimens between methods was detected. Age structure was different between samples from breeding sites and villages, with more gravid females collected in breeding sites, whereas more nulipars were collected in villages. By collection method, age structure was also different both in breeding sites and in villages. In breeding sites, the percentage of parous females was significantly higher in human landing catches, whereas the percentage of gravid females was significantly higher in light traps. In villages, only the percentage of gravid females was significantly higher in light traps. Our results suggests that UV light traps could be used to measure several entomological parameters of An. albimanus populations because both abundance variations and parity rates were similarly detected by both methods.

INTRODUCTION

Estimation of the abundance and age structure of vector populations has been of great importance in malaria epidemiology and vector control assessment (Macdonald 1957, Bruce-Chwatt 1985). In the context of malaria transmission, collection of mosquitoes with human bait provides a direct estimate of man-vector contact (World Health Organization 1975). However, the use of human bait is labor intensive, tedious, dangerous, uncomfortable, and expensive (Rubio-Palis and Curtis 1992), and the number of mosquitoes collected is affected by the ability of each collector and the variability in individual collectors' intrinsic attractiveness (Lindsay et al. 1993).

Several collection methods have been evaluated and compared with the human landing rate technique. These methods include resting collections, curtain traps, and various types of light traps and combinations of light traps with chemical attractants (Service 1977, Sexton et al, 1986, Rubio-Palis and Curtis 1992, Mbogo et al. 1993, Gunasekaran et al. 1994, Davis et al. 1995, Rubio-Palis 1996). Not all mosquito species are equally attracted to light traps. In the Americas, this method has been ineffective for collecting *Anopheles nuñeztovari* Gabaldon (Rubio-Palis and Curtis 1992) and *Anopheles pseudopunctipennis* Theobald (Fernández-Salas et al. 1994), but UV light traps have been successfully used to estimate *Anopheles albimanus* Wiedemann abundance (Sexton et al. 1986). Light trap collections could be standardized to allow comparisons among *An. albimanus* populations, but to date, there are no conclusive data about the age structure of the mosquitoes collected with light traps or how these collections compare with collections from human baits.

In order to choose a suitable collection method to estimate *An. albimanus* abundance in southern Mexico, we compared UV updraft light traps and human landing catches within villages and in nearby breeding sites. The results of this study and the age structure of these collections are reported herein.

MATERIALS AND METHODS

Study area: This study was conducted in 4 villages and 5 breeding sites distributed over an area approximately 49 km² of the southern Pacific coastal plain in the state of Chiapas (elevation 0-15 m), México. The villages were Emiliano Zapata (population 177) Barra San Simón (population 184), Efrain A. Gutiérrez (population 604), and Huanacaxtal (population 120) (Fig. 1). In general, houses in these villages were well ventilated, with palmthatch roofs and discontinuous walls made of palm poles, split bamboos, and, in some cases, cement block. Puente Baral is a breeding site at the edge of a mangrove area approximately 0.7 km from Emiliano Zapata. The main associations between An. albimanus larvae and vegetation in this breeding site were with Cynodon dactylon, Jouvea straminea, Lippia nodiflora, Batis maritima, and Cer-

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Fig. 1. Map of the study area.

atophyllum dermersun. El Paraíso breeding site is approximately 0.8 km from Barra San Simón. It is included in an estuary system irrigated by the Coatán River. During the dry season, when the sandbar is closed, the estuary becomes appropriate for associations between An. albimanus larvae and Crinun cf. erubescens, Paspalum vaginatum, Paspalum arundinacium, and Fimbristylis spadicea. El Burrero breeding site, approximately 1 km from Barra San Simón and 3 km from Efrain A. Gutiérrez, is considered a semipermanent lagoon tributary of the Coatan River. In this site, frequent associations of An. albimanus larvae occur with Paspalum sp., Brachiaria mutica, and Eichhornia crassipes. Zanjón Las Pitas breeding site is 2.1 km from Efrain A. Gutiérrez. It is a permanent lagoon fed by an intermittent stream. During the dry season, when the water flow decreases, larval habitats of Pistia stratiotes, Salvinia auriculata, Lemna minor, and Wolfia sp. are the most abundant. Huanacaxtal stream was in the past the original water course of the Coatán River and is 0.2 km from Huanacaxtal village. In this place, larval habitats are associated with Salvinia minima and Spirodella sp. (Rodríguez et al. 1993).

Mosquito collections: Adult mosquitoes were sampled at weekly intervals from January to April 1991. Light trap and human landing catches were simultaneously carried out in each breeding site and each village. After each collection day, a sample of 20 An. albimanus females from each collection method (or the whole collection when fewer than 20) was taken to the laboratory and dissected to determine parity (Detinova 1962). Mosquito identification was conducted according to the taxonomic key of Clark-Gill and Darsie (1983).

Breeding sites: In breeding sites, adult mosquitoes were sampled with 10 updraft UV light traps (John W. Hock, Gainesville, FL). Traps were located 20 m apart and 20 m from the water. Traps were operated from 1800 to 0600 h, after which mosquitoes were identified and counted. Human landing catches were carried out by 2 technicians during 50 min of each hour from 1800 to 2400 h. Collectors were fixed in stations next to the water body but at least 20 m away from the traps. Mosquitoes

	Mean number of An. albimanus collected ¹ Villages							
Collection method	SS	HU	EZ	EAG	Total	P (t-test)		
Human Landing	127.66 (166.78)	17.13 (21.84)	148.55 (105.81)	43.37 (55.41)	4,581	<u>. </u>		
Light trap	47.13 (65.23)	20.13 (20.17)	65.22 (70.81)	58.51 (107.43)	3,317	0.489		

Table 1. Average Anopheles albimanus adult abundance by collection method in villages and breeding sites.

¹SS = San Simón, Hu = Huanacaxtal, BU = Burrero, PA = Paraíso, EZ = Emiliano Zapata, EAG = Efraín A Gutiérrez, BA = Puente Baral, ZA = Zanjón Las Pitas.

attempting to bite were captured using mouth aspirators and placed in individual containers for each collection hour (World Health Organization 1975, Bown et al. 1987).

Villages: One trap operating from 1800 to 0600 h was placed indoors in each 1 of 3 houses selected in each village. Human landing catches were conducted indoors and outdoors by 2 people at the same time (but from 1800 to 2400 h) in 1 of the 3 houses. The collector was positioned on the opposite side of the house from where the light trap was placed.

Data processing: The total number of mosquitoes collected in all breeding sites and in all villages was pooled and the abundance expressed as the number of mosquitoes collected by 1 trap or 1 human bait and the total of specimens by method and by collection site (i.e., breeding site and village). A 1-way ANOVA test was used to test for differences between collection methods in villages and breeding sites. Relationships in numerical abundance of mosquitoes in villages versus nearest breeding sites were examined with correlation analyses. We used t-tests to compare catches between methods by collection site after log(X + 1) transformation. Regression analyses were used to determine the relationships between collecting methods by collection site after log(X + 1) transformations. Differences in parous rates between collection methods were examined by contingency table analyses and by ttests after arcsin transformations of percentages (Zar 1984).

RESULTS

Anopheles albimanus was the most abundant malaria vector caught in breeding sites and in villages. Other anophelines collected in light traps were Anopheles crucians Wiedemann, An. punctimacula Dyar and Knab, and An. vestitipennis Dyar and Knab. Anopheles punctimacula and An. vestitipennis were taken only in human bait collections. Because numerical data were compiled only for An. albimanus, the other species are not included in this report.

Villages with high An. albimanus abundance

were Emiliano Zapata and San Simón with human landing collections and Emiliano Zapata and Efrain A. Gutiérrez with light trap collections. Breeding sites with high abundance by both methods were El Burrero, Zanjon Las Pitas, and Puente Baral (Table 1). Significant differences in overall An. albimanus abundance among villages and among breeding sites were detected only by human landing catches (P < 0.005). Anopheles albimanus abundance in each village was not significantly correlated with its abundance at the nearest breeding site (P > 0.05).

Significantly more mosquitoes were captured in human landing collections in both villages and breeding sites $(34.3 \pm 6.3 \text{ and } 14.6 \pm 2.9 \text{ per hu-}$ man bait, respectively) than in light trap collections $(15.9 \pm 3.3 \text{ and } 2.4 \pm 0.3 \text{ per trap, respectively}; t$ = 2.206, df = 136, P = 0.0102; t = 4.105, df = 166, P = 0.0001). Also, the number of mosquitoes captured by both sampling methods was higher in villages (7,898) than in breeding sites (4,481; t = -4.501, df = 304, P = 0.0001).

After pooling, 2,465 An. albimanus females were collected in human landing collections and 2,016 by light trap collections in breeding sites. No significant differences were detected in abundance estimates by the 2 methods (t = 1.51, P = 0.133). There was significant correlation in numbers caught by the 2 methods Figure 2a). ($r^2 = 0.51$, P = 0.0001). In the villages, 4,581 An. albimanus were collected by human landing catches and 3,317 by light traps. The abundance ratio by the 2 methods in villages was similar to the abundance ratio observed in breeding sites (t = 0.69, P = 0.489). As with collections in breeding sites, a significant correlation was detected between the 2 methods ($r^2 = 0.785$, P = 0.0001; Fig. 2b).

A sample of 1,520 An. albimanus collected in breeding sites and another 1,637 collected in villages were dissected to determine parity (Table 2). Age structure was different between samples from breeding sites and villages ($\chi^2 = 276.79$, df = 2, P = 0.0001). In breeding sites, 29% were parous, 23% gravid, and 48% nulliparous; whereas in villages, 25% were parous, 4% gravid, and 71% nulliparous. When each age was compared between

Mean (±SD) number of An. albimanus collected								
Breeding sites								
BU	РА	HU	BA	ZA	Total	P (t-test)		
66.81 (95.82)	10.5 (12.34)	6.53 (6.86)	28.5 (31.6)	49.18 (54.53)	2,465	0.133		
23.43 (20.16)	21.25 (29.28)	18.96 (21.82)	26.3 (11.78)	34.93 (42.23)	2,016			

a) Breeding sites



Fig. 2. Numbers of *Anopheles albimanus* caught by light traps plotted against the number caught the same night by human landing collections in breeding sites (a) and villages (b).

Parity status	Breeding sites			Villages				Comparison between	
	Overall	Light trap	Human landing	P (t-test)	Overall	Light trap	Human landing	P (t-test)	and villages P (t-test)
n	1,520	971	549		1,637	446	1.191		
Parous (%)	29	23	39	0.0004	26	23	26	>0.05	>0.05
Gravid (%)	23	33	5	0.0001	4	10	20	0.0061	0.0001
Nulliparous (%)	48	44	56	>0.05	71	68	72	>0.05	0.0001

Table 2. Parity of Anopheles albimanus collected by two methods in breeding sites and in villages.

samples by collection site, significant differences were detected in the percentage of females that were gravid (t = 4.30, df = 127, P = 0.0001) and nullipars (t = -4.81, df = 126, P = 0.0001).

The age structure of mosquitoes was different between sampling methods, both in breeding sites $(\chi^2 = 166, df = 2, P = 0.0001)$ and in villages (χ^2) = 46.6, df = 2, P = 0.0001). In breeding sites, light traps collected 23% parous, 33% gravid, and 44% nulliparous females, whereas females from human landing collections were 39% parous, 5% gravid, and 56% nulliparous. When each age group was compared between methods, only the percentages of females that were parous (t = 3.64, df =156, P = 0.0004) and gravid (t = -5.97, df = 16, P = 0.0001) were significantly different. In villages, light trap samples were 23% parous, 10% gravid, and 67% nulliparous, whereas human landing samples were 26% parous, 2% gravid, and 72% nulliparous. When each age was compared between methods, significant differences were detected only in the percentage of females that were gravid (t =-2.78, df = 126, P = 0.0061).

DISCUSSION

Anopheles albimanus is an exophilic and zoophagic malaria vector in Mexico and Central America (Rodríguez and Loyola 1989), with extremely low sporozoite rates (Ramsey et al. 1994). Malaria transmission efficiency of this mosquito is highly dependent on overall population abundance, which peaks during the rainy season (Bown et al. 1991). On the other hand, when control methods, such as indoor insecticide spraying, are evaluated, the depletion of the older portion of the population is a better indicator of control efficacy than is overall vector abundance (Bown et al. 1991). Consequently, it is important that sampling methods provide a good estimate of vector abundance and an unbiased estimate of vector age.

In a previous survey in Haiti, Sexton et al. (1986) demonstrated that UV light traps were more effective than human biting collections for sampling *An. albimanus* adults, both outdoors and indoors. In our study, regardless of collection site (village or breeding site) and in spite of human landing collections being conducted for 6 h versus whole night collections for light traps, human landing collections captured more mosquitoes per night. No differences in the way the studies of Sexton et al. (1986) and our studies were performed explain these contradictory results. Light traps also captured more *Anopheles* (*Cellia*) gambie Giles s.1. and *Anopheles* (*Cellia*) funestus Giles than did human bait in Tanzania (Davis et al. 1995) but were less attractive to the same species in Kenya (Mbogo et al. 1993). These discrepancies may be explained by factors such as variation in host preference, feeding behavior, and seasonality (Davis et al 1995).

In our study, the abundance ratios for the 2 sampling methods were not significantly different in the 2 collection sites, but a significant correlation between the number of mosquitoes collected in light traps and human landing collections was observed, indicating that abundance variations detected in human collections were similarly detected by trap collections. Different studies with several anopheline species such as *An. nuñeztovari, An. triannulatus* Neiva and Pinto, *An. albitarsis,* and *An. oswaldoi* Peryassu from Venezuela (Rubio-Palis and Curtis 1992) and *An. gambiae* (s.l.) and *An. funestus* from Kenya (Mbogo et al. 1993) have also demonstrated that the average catches using light traps and human bait were statistically similar.

Independent of collection method, the age structure of *An. albimanus* females was different between breeding sites and villages. Specifically, the percentages of females that were gravid and nulliparous were significantly different between the 2 sites. The greater number of females in a gravid state in breeding sites may correspond to mosquitoes returning to oviposit, whereas the greater number of nulliparous females in villages is a results of a large number of females seeking their first blood meal. These differences allowed us to make comparisons on the performance of the collection methods in 2 different portions of the same population.

Rubio-Palis and Curtis (1992) reported high parity rates in human landing versus light trap collections for *An. nuñeztovari* but not for *An. albitarsis* and *An. triannulatus* collected in villages. This indicates that attractiveness to light traps may vary significantly among species. In our study, light traps were more sensitive to the presence of gravid *An. albimanus* than landing captures, both in breeding sites and in villages. This may be indicative, not of the intrinsic attractiveness of the device to the mosquito but of the effect of the physiological state on the attraction of mosquitoes to humans. Human landing catches collected proportionally more parous females than light traps in breeding sites but not in the villages. This result could be a consequence of the bias introduced by the high numbers of gravid females (attracted to light traps) in breeding sites. However, when gravid females were eliminated from the samples, the proportion of parous females collected by human landing collections remained significantly higher (P = 0.0204) than those collected by light traps. If we consider that mosquitoes attracted to humans, no matter the location (breeding site or villages), are displaying a blood-seeking behavior and that An. albimanus is a zoophilic mosquito (with strong preference for cows (Arredondo-Jimenez et al. 1992), the lower proportion of parous females attracted to humans in villages could be explained by the availability of other vertebrate hosts (undetermined numbers of cows, horses, and pigs). In breeding sites, no large mammals were observed, and the only vertebrate host was represented by the human baits. We might speculate that An. albimanus females in villages might selectively opt for vertebrate hosts other than humans.

In summary, our results support the use of light traps to measure several entomological parameters of An. albimanus populations. Variations in abundance were detected similarly by both collection methods. Similar parity rates were also detected by both methods, which makes the use of light traps suitable for the evaluation of survival rate. longevity, and sporozoite rate, as well as for assessments of the impact of vector control measures. Light traps have the additional advantage over conventional capture methods of being more cost effective because of decreased human resource costs (with only 2 people, it is possible to monitor at least 14 villages with 4 traps per village on a weekly basis; Beck et al. 1994, 1997; Rodríguez et al. 1996) and more ethical because collection personnel are no longer at risk of acquiring infection (as human bait).

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