Medical and Veterinary Entomology (2008) **22**, 120–123

The efficacy of indoor CDC light traps for collecting the sandfly *Phlebotomus argentipes*, vector of *Leishmania donovani*

D. S. DINESH[1], P. DAS[1], A. PICADO[2], C. DAVIES[2], N. SPEYBROECK[3,4], M. BOELAERT[5] and M. COOSEMANS[6,7]

[1]Rajendra Memorial Research Institute of Medical Sciences, Indian Council of Medical Research, Agamkuan, Patna, India, [2]Department of Infections and Tropical Diseases, London School of Hygiene and Tropical Medicine, London, U.K., [3]Department of Animal Health, Institute of Tropical Medicine, Antwerp, Belgium, [4]Ecole de Santé Publique, Université Catholique de Louvain, Bruxelles, Belgium, [5]Department of Public Health, Institute of Tropical Medicine, Antwerp, Belgium, [6]Department of Parasitology, Institute of Tropical Medicine, Antwerp, Belgium, [7]Department of Biomedical Sciences, Faculty of Pharmaceutical, Veterinary and Biomedical Sciences, University of Antwerp, Belgium.

Abstract. The efficacy of light traps for collecting sandflies (Diptera: Psychodidae) varies both inter-specifically and intra-specifically (by gender and physiological status) as a result of significant differences in phototropic and other behavioural characteristics. The efficacy of miniature CDC light traps for collecting *Phlebotomus argentipes* Annandale & Brunetti, a vector of *Leishmania donovani* Laveran & Mesnil (Kinetoplastida: Trypanosomatidae), was assessed in the Indian state of Bihar. Sandflies were collected during the night from 16 houses in each of three villages over 3 months (four times at fortnightly intervals) using CDC light traps indoors, and by aspirator collection (carried out by one person for 30 min/house) from the walls of the same houses the following morning. Incidence rate ratios (IRRs) between CDC light trap collections and aspirator collections were obtained through a negative binomial regression with household as random effect. CDC light traps were especially effective in catching males (IRR 3.08, 95% confidence interval [CI] 2.12–4.46) and unfed females (IRR 3.50, 95% CI 2.37–5.16) of *P. argentipes*, and to a lesser extent gravids (IRR 1.77, 95% CI 1.07–2.93). However, only a relatively small proportion of all blood-fed *P. argentipes* were collected by light trap (IRR 0.45, 95% CI 0.28–0.73). Despite its limitations in collecting blood-fed female sandflies, the CDC light trap appears to trap a sufficient proportion of the indoor population of sandflies for sampling purposes, and as this light trap is also more convenient and more easily standardized than the aspirator method, we conclude that it is the most efficient method for monitoring *P. argentipes* populations in the Indian subcontinent.

Key words. *Phlebotomus argentipes*, CDC light trap, sampling methods, sandflies, trapping, Bihar, India.

Introduction

Visceral leishmaniasis, caused by *Leishmania donovani* Laveran and Mesnil, is a major health problem for people in the Indian states of Bihar, Uttar Pradesh, West Bengal and Jharkhand. *Phlebotomus argentipes* Annandale and Brunetti, which is both endophagic and endophilic (Hati, 1991), is the proven vector of *L. donovani* (Swaminath *et al.*, 1942; Kumar *et al.*, 2001). A variety of methods for collecting sandfly species have been used for research purposes and the evaluation of vector control programmes, particularly sticky traps, aspirators and light traps. Sticky traps, which are limited by their surface area (20×29 cm),

Correspondence: Marc Coosemans, Department of Parasitology, Institute of Tropical Medicine, Nationalestraat 155, B-2000 Antwerp, Belgium. Tel.: + 32 32476312; Fax: + 32 32476359; E-mail: mcoosemans@itg.be

capture predominantly males and unfed females, and to a lesser extent females of other physiological statuses (Kaul *et al.*, 1994a). Light traps are less labour-intensive, but their efficacy depends on the phototropic behaviour of the target species (Davies *et al.*, 1995). To our knowledge, there have been no previous studies designed to evaluate the efficacy of light traps for collecting sandflies in the Indian subcontinent, and most previous studies or surveys of *P. argentipes* in this region have depended on sticky traps or direct aspiration (Kaul *et al.*, 1994b; Kumar *et al.*, 1995; Ghosh *et al.*, 1999; Palit *et al.*, 2005). In this study, the relative efficacy of the CDC (Centers for Disease Control and Prevention) light trap was assessed by determining the incidence rate ratios (IRRs) of sandflies collected in houses by light traps vs. aspirators. These ratios were stratified by sex and physiological status.

Materials and methods

Sandfly collection

Three visceral leishmaniasis-endemic villages in the state of Bihar, India were selected for the study: Gulmehiya Bagh (Patna district), and Rasoolpur and Majlishpur (Vaishali district). In each village, 16 houses were selected as being representative of village dwellings. All had earthen floors, tiled or thatched roofs and brick, dried mud or thatched walls. Four consecutive sandfly surveys were performed in each village, at fortnightly intervals from April to June 2006, corresponding to the main sandfly season (Dinesh *et al.*, 2001). During each village visit, sandflies were collected by miniature CDC light traps (miniature incandescent light trap, model 1012; J. W. Hock Co., Gainesville, FL, U.S.A.) from one room in each of the selected houses between 18.00 hours and 06.00 hours. This was followed by an aspirator collection carried out by one trained person for a 30-min period between 06.00 hours and 08.00 hours the following morning in all of the two or three rooms of the same house, including the bedroom where the CDC light trap had been placed. Informed consent was obtained from the head of each household. The CDC light traps were hung near the corner of a bedroom, 6 inches above the ground and 1 inch away from the wall. In each bedroom sleepers were protected by bednets. The labelled pots that held sandflies were detached from the traps in the morning and the sandflies were released into a Baraud cage. Dead sandflies were picked up using a moistened camel hair brush and put in a labelled Petri dish. Live sandflies were killed by keeping them in the refrigerator for 1 hour. The sandflies collected by aspirator were released in test tubes and transported to the laboratory to be processed in the same way. All sandflies were examined under a binocular dissecting microscope for identification.

Statistical analysis

A negative binomial model was used because of overdispersion in the count data. A random-effects model was used because samples from within a household shared the same environment (physical location), as well as specific characteristics (Speybroeck *et al.*, 2003). These shared factors, the effects of which can change from household to household, create dependencies between responses observed for individual collections (e.g. CDC light trap or aspirator collections). The 'xtnbreg' command in Stata 9 (StataCorp LP, College Station, TX, U.S.A.) was used to fit the random-effects negative binomial regression models.

Results

A total of 2462 sandflies was collected from April to June 2006. Of these, 1655 (67.3%) were collected by CDC light trap and 807 (32.8%) by aspirator (Table 1). Sandflies belonging to two genera (*Phlebotomus* and *Sergentomyia*; 1047 and 1415, respectively) were captured during the four surveys. Both genera were collected in greater numbers by CDC light traps (30.5% and 36.8%, respectively, of all collected sandflies) than by aspiration (12.1% and 20.7%, respectively). Nearly all the *Phlebotomus* sandflies collected were *P. argentipes* (1039). The remaining eight were *Phlebotomus papatasi*.

The light traps performed well in attracting males and unfed females of both *P. argentipes* (81.7% and 80.8%, respectively, of the totals collected by either method) and *Sergentomyia* sp. (71.2% and 67.3%, respectively) (Fig. 1). The negative binomial model shows that, when controlling for the possible differences

Table 1. Numbers and percentages of sandflies by sex and physiological status, collected overnight by CDC light traps followed by early morning aspirator collections.

	Phlebotomus argentipes						*Sergentomyia* spp.						
	Male	Female					Male	Female					
CM		UF	F	Gr	ST	T		UF	F	Gr	ST	T	Totals*
CDC %	14.9	10.4	1.5	3.4	15.3	30.2	13.8	18.2	1.5	3.2	22.9	36.8	67.2
ASP %	3.3	2.5	4.3	1.8	8.6	12.0	5.6	8.8	3.4	2.9	15.1	20.7	32.8
Total	449	317	144	129	590	1039	479	664	120	152	936	1415	2462
n (%)	(18.2)	(12.9)	(5.8)	(5.2)	(24.0)	**(42.2)**	(19.5)	(27.0)	(4.9)	(6.2)	(38.0)	**(57.5)**	

*Totals include eight specimens of *Phlebotomus papatasi*.
CM, collection method; ASP, aspiration; CDC, CDC light trap; UF, unfed; F, fed; Gr, gravid; ST, subtotal (i.e. of females); T, total of females and males.

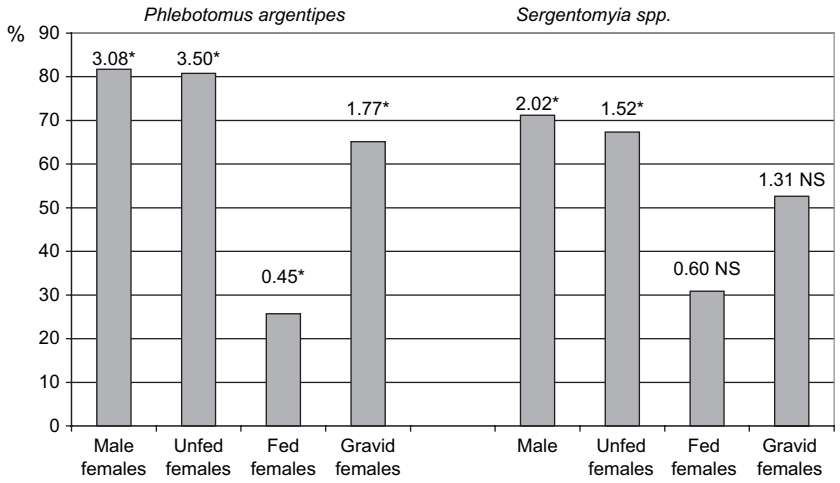

Fig. 1. Yields in *Phlebotomus argentipes* and *Sergentomyia* spp. by sex and physiological status of CDC light trap collections as a percentage of total collected sandflies per category. (Total collected sandflies comprised those collected by CDC light traps during the night and those collected by aspirators in the morning.) Incidence rate ratios are reported for each group (*$P < 0.05$; NS, not significant).

between surveys, light trap collections provided about three times as many males (IRR 3.08, 95% confidence interval [CI] 2.12–4.46) and unfed females (IRR 3.50, 95% CI 2.37–5.16) of *P. argentipes* than with the subsequent aspiration method. A similar trend was observed with male and unfed *Sergentomyia* spp. (IRR 2.02, 95% CI 1.48–2.76 and IRR 1.52, 95% CI 1.14–2.04, respectively) and gravid females of *P. argentipes* (IRR 1.77, 95% CI 1.07–2.93). By contrast, CDC light traps provided less than half of all blood-fed *P. argentipes* collected (IRR 0.45, 95% CI 0.28–0.73) (Table 2). No significant differences were observed between the first survey and the other three surveys, except for fed females of *Sergentomyia* spp. (results not shown).

Discussion

Reliable data from vector collection surveys are crucial to research and for the evaluation of vector control programmes. Several studies in the past have tried to assess the different methods available to collect *Phlebotomus*. An 18-year retrospective study carried out in Italy showed that CDC light traps were more effective in collecting *Phlebotomus perfiliewi*, the predominant species in the domestic environment in Italy, than sticky traps, window exit traps, emergence traps, or hand catches from domestic and sylvatic habitats in rural, urban and periurban areas (Maroli *et al.*, 1994). A study on the natural behaviour of sandflies in Egypt (South Sinai and Alexandria) adopted various sampling methods, including CDC light traps, aspirators, funnel traps, fan traps, catches of baits, sticky traps and illuminated sticky traps. Comparisons between the efficacies of the different trapping methods for collecting sandflies indicated that the CDC light trap was the most productive for species of the genus *Phlebotomus*, followed by illuminated paper traps and sticky traps (Hilmy *et al.*, 1989).

Prior to this study, the data available from the Indian subcontinent were limited to the results of a pilot study to evaluate different sandfly sampling methods conducted in 2005 in two different leishmaniasis-endemic villages in Bihar (India), in which the CDC light trap performed better than the CDC ultraviolet trap, the sticky trap or the aspirator (D. S. Dinesh, unpublished data, 2005). In the present study, the performance of the CDC light trap was assessed by examining the resting population collected in the morning using the aspirator method after the light trap collection had taken place. This assumes that the successive collection methods together provide a representative sample of the population of sandflies inside the houses. The

Table 2. Incidence rate ratios of CDC light trap collections vs. subsequent aspirator collections obtained through a negative binomial regression with household as a random effect.

	Phlebotomus argentipes				*Sergentomyia* spp.			
	Males	Unfed females	Fed females	Gravid females	Males	Unfed females	Fed females	Gravid females
CDC vs. aspirator	3.0783	3.4963	0.4533	1.7742	2.0199	1.5241	0.5979	1.3093
Lower CL	2.1225	2.3670	0.2833	1.0743	1.4782	1.1386	0.3465	0.8211
Upper CL	4.4647	5.1643	0.7252	2.9300	2.7600	2.0402	1.0316	2.0879
P-value	>0.0001	>0.0001	0.0010	0.0250	>0.0001	0.0050	0.0650	0.2580
Nbreg-parameters								
R	3.2930	12.7422	3.9648	3.3900	8.3160	4.0520	2.5977	24.4844
S	7.6036	24.1688	2.6906	2.6571	21.9079	12.8357	2.6428	53.1448

Note: The inverse of one plus the dispersion is assumed to follow a beta (r,s) distribution. In all models the panel estimator was significant compared with the pooled estimator (i.e. a negative binomial estimator with constant dispersion).
Lower CL, lower limit of 95% confidence interval; Upper CL, upper limit of 95% confidence interval.

overall sex ratio for *P. argentipes* collected by both methods was 0.76, which is not very different from the 0.83 obtained by the aspiration method used alone during the monsoon (Ghosh *et al.*, 1999). Resting site collections such as by direct aspiration usually provide a good picture of the overall fauna (Alexander, 2000) and can here be used to estimate the relative efficacy of the CDC trap. The CDC light trap collection method was found to be effective in sampling *P. argentipes*, particularly for males and unfed females, and, to a lesser extent, gravid females, but not blood-fed females. No controls were used in present study (e.g. we did not use the aspirator method in houses where no CDC trap was used overnight) and thus we cannot conclude that CDC light traps are more effective than the aspirator method. However, the aspirator method is more labour-intensive: one person can cover a maximum of three houses in one morning (06.00−08.00 hours), but can collect CDC traps from more than 20 houses in the same time. Moreover, large differences in the aptitudes of collectors make it difficult to standardize the aspirator method (Alexander, 2000), whereas this is easily achieved by specifying the location of CDC traps. One CDC light trap (including battery and charger) costs about US$300 and lasts for about 5 years with a minimum of maintenance.

Given that the CDC light trap method is less labour-intensive and thus cheaper, that it is more easily standardized than the aspirator method and that it catches $>60\%$ of collectable males and unfed females, we conclude that it is the most effective method available.

The CDC light trap is useful in measuring relative changes in abundance but has limited value in ecological studies (Alexander, 2000). To improve the representativeness of samples, we need to acquire more knowledge of the behaviour of blood-fed females as neither method was very successful in collecting this physiological stage. The CDC light trap has already been shown to be an effective and suitable tool for monitoring leishmaniasis control programmes in Peru (Davies *et al.*, 1995). The results from this study show that a similar approach can be adopted in Bihar and, perhaps, in other parts of India.

Acknowledgements

The authors are thankful to the European Union for providing financial support (proposal contract no. 015374); the Indian Council of Medical Research for providing facilities and obtaining approval from the Government of India Health and Family Welfare Department, New Delhi; the staff of the Division of Vector Biology and Control, Rajendra Memorial Research Institute of Medical Sciences of Patna and the KALANET project staff for laboratory and field assistance. The purchase of CDC light traps was sponsored by G. Deckers (Konhef, VZW, Antwerp, Belgium).

References

Alexander, B. (2000) Sampling methods for phlebotomine sandflies. *Medical and Veterinary Entomology*, **14**, 109–122.

Davies, C.R., Lane R.R., Villaseca, P., Pyke, S., Campos, P. & Llanos-Cuentas, A. (1995) The relationship between CDC light trap and human bait catches of endophagic sandflies (Diptera: Psychodidae) in the Peruvian Andes. *Medical and Veterinary Entomology*, **9**, 241–248.

Dinesh, D.S., Ranjan, A., Palit, A., Kishore, K. & Kar, S.K. (2001) Seasonal and nocturnal landing/biting behaviour of *Phlebotomus argentipes* (Diptera: Psychodidae). *Annals of Tropical Medicine & Parasitology*, **95**, 197–202.

Ghosh, K., Mukhopadhyay, J., Desai, M., Senroy S. & Bhattacharya, A. (1999) Population ecology of *Phlebotomus argentipes* (Diptera: Psychodidae) in West Bengal, India. *Journal of Medical Entomology*, **36**, 588–594.

Hati, A.K. (1991) A review of *Phlebotomus (Euphlebotomus) argentipes* Annandale & Brunetti - the vector of Kala-azar (visceral leishmaniasis) in India. *Indian Review of Life Sciences*, **11**, 53–75.

Hilmy, N.M., Shehata, M.G., el Housary, S., Kamal, H., Doha, S. & el Said, S. (1989) Investigation of sampling methods for the study of Phlebotomine sandflies. *Journal of the Egyptian Public Health Association*, **64**, 401–415.

Kaul, S.M., Khera, A.K. & Bora, D. (1994a) A comparative study on the optimum time and method of collection of *Phlebotomus argentipes* and other sandflies (Diptera: Psychodidae). *Journal of Communicable Diseases*, **26**, 172–176.

Kaul, S.M., Sharma, R.S., Borgohain, B.K., Das, N.S. & Verghese, T. (1994b). Absence of *Phlebotomus argentipes* Ann & Brun. (Diptera: Psychodidae), the vector of Indian Kala-azar from Kamrup District, Assam. *Journal of Communicable Diseases*, **26**, 68–74.

Kumar, V., Kesari, S.K., Sinha, N.K. *et al*. (1995) Field trial of an ecological approach for the control of *Phlebotomus argentipes* using mud and lime plaster. *Indian Journal of Medical Research*, **101**, 154–156.

Kumar, V., Kishore, K., Palit, A. *et al*. (2001). Vectorial efficacy of *Phlebotomus argentipes* in Kala-azar endemic foci of Bihar (India) under natural and artificial conditions. *Journal of Communicable Diseases*, **33**, 102–109.

Maroli, M., Bigliocchi, F. & Khoury, C. (1994) Sandflies in Italy: observations on their distribution and methods for control. *Parassitologia*, **36**, 251–64.

Palit, A., Bhattacharya, S.K. & Kundu, S.N. (2005) Host preference of *Phlebotomus argentipes* and *Phlebotomus papatasi* in different biotopes of West Bengal, India. *International Journal of Environmental Health Research*, **15**, 449–545.

Speybroeck, N., Boelaert, F., Renard, D., Burzykowski, T., Mintiens, K., Molenberghs, G. & Berkvens, D.L. (2003) Design-based analysis of surveys: a bovine herpesvirus 1 case study. *Epidemiology and Infection*, **131**, 991–1002.

Swaminath, C.S., Shortt, H.E. & Anderson, L.A.P. (1942) Transmission of Indian Kala-azar to man by bites of *Phlebotomus argentipes* Ann and Brun. *Indian Journal of Medical Research*, **30**, 473–477.

Accepted 14 February 2008

