EVALUATION OF A FUNNEL TRAP FOR COLLECTING COPEPODS AND IMMATURE MOSQUITOES FROM WELLS

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ABSTRACT. A simple funnel trap which inverts on entry to and exit from the water, was evaluated with respect to sampling the aquatic fauna of wells in and around Fortaleza, Brazil. Mosquito larvae, copepods, ostracods, fish and tadpoles were collected. When known numbers of *Culex quinquefasciatus* larvae and *Mesocyclops aspericornis* copepods were introduced into negative wells, overnight sampling resulted in 50% positivity of traps when 10 larvae (1/1,000 cm^2) and 25 copepods (1/176 liters) were introduced. All traps were positive when 100 and 400 larvae and copepods, respectively, were present. Over a range of 10–2,000 and 25–800 released larvae and copepods, respectively, average recovery rates were 6.1 and 3.6%, but on the basis of one sample, the variability was such that a precise estimate of absolute abundance could not be made.

INTRODUCTION

Service (1976) has reviewed a variety of dippers, weighted cone-shaped nets, and plastic and metal cylinders with flaps used for sampling immature mosquitoes breeding in wells. In Myanmar (Burma), Tun-Lin et al. (1988) found their modified netting procedure to be superior to that recommended by the World Health Organization (1975) and reported limitations in the latter method for well water surfaces greater than 3 m below ground level. The modified method involved immersing a weighted net 60 cm below the well water surface and after a minute, drawing it up through marginal debris. In Brazil, the current method is to drop a bucket into the well 3 times and to inspect the retrieved water for immatures.

The funnel trap described herein is essentially a simplification of earlier traps used for collecting *Culiseta melanura* (Coq.) from ground holes in Maryland (Muul et al. 1974) or for *Aedes aegypti* (Linn.) and *Culex quinquefasciatus* Say in water storage pots in Thailand (Harrison et al. 1982). Our problem in Ceará, Brazil involved the efficient monitoring of these 2 latter species in relation to numbers of copepods released for biocontrol. Wells are common in rural villages and the water surface may be 10–15 m below ground level, thus making sampling difficult.

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MATERIALS AND METHODS

Funnel trap design: The trap functioned on 2 premises: 1) immature mosquitoes and copepods were guided by the inverted funnel into the reservoir when they swam toward the surface, and 2) trapping of air in the reservoir ensured that the device floated unaided. It comprised 4 parts: 1) a 22 cm diam. plastic funnel, 2) a 500 ml polystyrene jar (reservoir) with screw cap (Corning cat. no. 25628-500), 3) a 50 g sinker, and 4) cord for attachment with a length, depending on the depth of the wells (Fig. 1). Spare reservoirs and caps were useful for taking the catch back to the laboratory. When the reservoirs were removed, the funnels (plus cord and sinker) stacked conveniently on top of each other.

The trap was assembled by cutting out the middle of the screw cap along the inside of the concentric groove and glueing this ring onto the funnel spout approximately 5–6 cm below its tip. The cord was knotted before and after threading through a hole at the rim of the funnel, leaving 10 cm spare for sinker attachment. Double knotting prevented the sinker from pulling the attachment cord taut so that the trap could float unhindered, and loss of the trap if the sinker became detached.

To operate, the jar was filled with 400 ml clean water which when inverted left a 3–4 cm air pocket for flotation. The jar was then screwed onto the funnel and the trap lowered, reservoir-up, into the well. On immersion, the weight of the sinker inverted the trap into its collecting mode. For retrieval, the trap was inverted by pulling the cord, coming out of the well funnel-up.

Evaluation of the trap: Trap efficiency was compared by day and by night within the Federal University of Ceará grounds in a 1,000 liter *Mesocyclops aspericornis* (Daday) mass culture tank and in 1.5 × 1 × 0.3 m sections of an

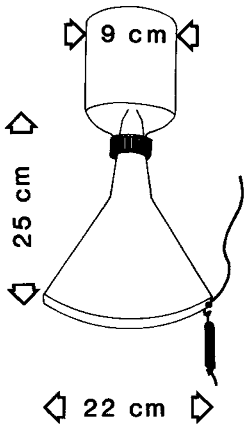

Fig. 1. The funnel trap: from top; reservoir, funnel and sinker.

effluent treatment pond containing *Cx. quinquefasciatus*. Over six 24-h periods, one and 2 traps for *M. aspericornis* and *Cx. quinquefasciatus*, respectively, were used over the hours of 1830–0715 and 0730–1800. Contents of each reservoir were emptied into white trays for counting and then returned to their respective sites. Although the traps were moved to different positions within each site, the integrity of the paired day vs night comparisons was preserved. Log (x + 1) counts were then compared by Student's *t*-test.

To determine if immature *Cx. quinquefasciatus* were escaping from the reservoir through the 2 cm diameter funnel tip, 150–200 third-fourth instar larvae were put in the reservoir and those remaining were counted after 10 hours. This was repeated with a reduced nozzle diameter of 1 cm with both *Cx. quinquefasciatus* and *M. aspericornis*.

The suburb of Granja Portugal in Fortaleza was chosen for survey because of its numerous wells. From February to April 1992, 285 wells were surveyed for immature mosquitoes, fish and copepods by trapping from approximately 1700 to 0800 h. Surface area, well depth and water volume were also recorded. In order to determine the sensitivity and efficacy of the funnel traps, the following numbers (and number of replicates) of mixed third-fourth instar *Cx. quinquefasciatus* were introduced into negative wells: 10 (8), 25 (12), 50 (8), 100 (9), 200 (8), 400 (6), 600 (4), 1000 (3), 1500 (6) and 2000 (4). From 0800 to 1700 h prior to each of these 68 releases, traps were left in the wells to recheck their negativity.

On the next day the following numbers (and number of replicates) of *Mesocyclops aspericornis* were released into the wells: 25 (4), 50 (4), 100 (4), 200 (4), 400 (4), 500 (2) and 800 (4). As before, the traps were run from 1700 to 0800 h and the number recovered counted in white trays. The numbers recovered in the traps were then analysed in relation to numbers released using SPSS statistics software.

RESULTS

Time of trapping: During night and day, respectively, the following totals were collected: *Cx. quinquefasciatus* larvae 501 and 299, pupae 61 and 24; *M. aspericornis* 3,860 and 3,066. When log (x + 1) catches were compared by *t*-test, there were no significant differences either for *Cx. quinquefasciatus* ($t = 1.18$, df = 11, $P = 0.26$) or for *M. aspericornis* ($t = 0.34$, df = 5, $P = 0.75$).

Escape from the trap: After 6 × 12 h trapping periods, from 12.1 to 29.3% of *Cx. quinquefasciatus* larvae had swum out of the reservoir. With a reduced opening of 1 cm diameter, losses were still of the same order. During 3 overnight trials with the 1 cm diameter nozzle and *M. aspericornis*, 8–12% had escaped from the reservoir by the next morning.

Sensitivity: The surface area of the funnel in comparison with that of the wells was 3.8%. The overall average recovery for *Cx. quinquefasciatus* and *M. aspericornis*, respectively, was 6.1 and 3.6%. For *Cx. quinquefasciatus*, percent recovery tended to decrease with increasing population size. The sensitivity of the overnight funnel traps for detecting presence of *Culex* larvae or *Mesocyclops* was dependent on the number of animals released. Detection success reached 100% for 100 or more larvae (Table 1) or 400 or more copepods (Table 2). The lowest sensitivity noted was 50% of traps positive for 10 larvae or for 25 copepods. At this level of infestation, repeated trappings would increase the eventual sensitivity of detection to the following levels: 75, 87, 94 and 97% success for the second, third, fourth and fifth nights, respectively.

Prediction: Pearson's correlation coefficients of $r = 0.84$ and 0.73 for copepods and larvae, respectively, indicated that the proportion recovered was relatively constant across a range of population levels (Tables 1 and 2). From this, r^2 values suggested that 71% and 53% of variation in the numbers captured were explained by variation in the numbers of copepods and larvae released. However, on the basis of one trap night per well, interpolation of catch size to the size of the natural population would be imprecise. Thus, regression equations to predict populations on the basis of the number captured have not been included.

Survey of wells: For 285 wells in Granja Portugal, the mean surface area ± SD (range) and mean distance from the well rim to the water

Table 1. Mean recovery of *Culex quinquefasciatus* larvae from funnel traps placed overnight in wells, in relation to number of larvae released.

No. released	No. trials	Recovery: Mean no. ± SD	Recovery: Range	Recovery: %	Sensitivity (% positive)
10	8	0.8 ± 1.0	0–3	7.5	50
25	12	1.9 ± 1.6	0–5	7.7	83
50	8	1.6 ± 1.7	0–4	3.3	63
100	9	6.9 ± 1.5	6–9	6.9	100
200	8	18.9 ± 8.2	11–36	9.4	100
400	6	19.2 ± 8.0	9–32	4.8	100
600	4	40.3 ± 12.1	23–50	6.7	100
1,000	3	46.3 ± 9.3	36–54	4.6	100
1,500	6	35.7 ± 12.4	14–47	2.4	100
2,000	4	81.5 ± 75.0	30–193	4.1	100
Totals	68			6.1	87

Table 2. Mean recovery of *Mesocyclops aspericornis* from funnel traps placed overnight in wells, in relation to number of copepods released.

No. released	No. trials	Recovery: Mean no. ± SD	Recovery: Range	Recovery: %	Sensitivity (% positive)
25	4	0.5 ± 0.6	0–1	2.0	50
50	4	2.5 ± 3.1	0–7	5.0	75
100	4	3.8 ± 2.9	0–6	3.8	75
200	4	3.0 ± 3.8	0–8	1.5	50
400	4	20.5 ± 19.2	3–46	5.1	100
500	2	17.5 ± 7.8	12–23	3.5	100
800	4	35.5 ± 7.0	31–46	4.4	100
Totals	26			3.6	77

level ± SD (range) was 1.1 ± 0.35 m^2 (0.7 – 4.3 m^2) and 1.5 ± 1.1 m (0 to 6.3 m). Well volume ranged from 4,400 to 11,600 liters. No wells were positive for *Ae. aegypti*; 27 were positive for *Cx. quinquefasciatus*, 35 for cyclopid copepods including *Mesocyclops* spp. and 59 for fish. The total number of larvae and pupae collected, respectively, was 9,902 and 1,055. In one heavily infested well, the funnel trap contained ≥5,000 larvae and ≥500 pupae after one night's trapping. After eliminating this extreme catch from calculations, the mean numbers of larvae and pupae recovered from the funnel traps were 188.5 and 17.5, respectively.

DISCUSSION

Apart from the evaluations at Granja Portugal where water levels were high and free of debris, we have also used the funnel traps successfully in wells at Vila Manuel Satiro and at Preaoca where the water surface was 8–10 m below ground level and sometimes littered with floating coconuts and wood. We have collected mosquito immatures, copepods, ostracods, fish and tadpoles.

After approximately 350 trap nights, *Ae. aegypti* has yet to be collected from wells in Ceará. However, approximately 10% contained *Cx. quinquefasciatus* immatures and in some cases in enormous numbers. Harrison et al. (1982) considered their AFRIMS (Armed Forces Research Institute of Medical Sciences) trap to be more efficient for collecting *Cx. quinquefasciatus* than *Ae. aegypti* because of their comparative feeding habits. Our trap would have similar characteristics to the AFRIMS trap but contains less parts. With one sinker, the inverted funnel lies in the water approximately 5° off the horizontal, but this is inconsequential. A second sinker can be added if desired but cost is a major consideration when working in developing countries. This model costs US $4 and can be used in water depths over 40 cm. This design could be produced in different sizes (and with different and less expensive materials) to accommodate mosquito and copepod biotopes of different sizes and depths. The important developmental considerations are the trapping of air within the reservoir and the balance achieved between the sinker and reservoir of water to make the trap invert on immersion.

Although efficiency seemed similar for day and night, trapping by night was more practical as the water surface was less likely to be disturbed through water retrieval for domestic usage. Consequently, trap damage was reduced. Trap returns of *Cx. quinquefasciatus* and *M. aspericornis* averaged 6.1 and 3.6%, respectively, but on the basis of a single night, the variability in returns precluded precise estimation of population size. Although 53% of the variability with the returns of *Cx. quinquefasciatus* was explained by population size, catches could be influenced the position of the trap (i.e., against the side or in the middle) and by clustering of the larval population itself. We are not convinced that the apparent tendency for reduced percent recovery of *Cx. quinquefasciatus* with increased population size is real although the clustered distribution may change when the population is large.

As the modes of respiration of mosquito larvae and copepods are different, we would expect the funnel traps to be more sensitive for detecting the former. For *Cx. quinquefasciatus* larvae, 50% were positive at 1 larva/1,000 cm^2 (10 per well) and all were positive at 1/100 cm^2 (100 per well). As *Mesocyclops aspericornis* occupies all of the water column but primarily is benthic (Brown and Kay, unpublished data), the detection of 25 in a well containing a minimum of 4,400 liters of water is particularly pleasing (1/176 liters).

Sensitivity was a key issue with respect to development of these funnel traps for sampling from wells. Currently, all wells are treated with temephos granules at 1 g/liter as part of national health policy. For the 285 wells surveyed at Granja Portugal, this implies usage of approximately 2,000 kg per treatment. If policy is altered to treat only the 27 wells found positive then less than 200 kg would be required. Should greater precision be required as a policy base, then trapping over 4 consecutive nights per negative well yields 95% confidence of less than 10 larvae/well. On the basis of laboratory evaluation (Kay et al. 1992), we intend to monitor the efficacy and persistence of predacious cyclopids, especially *M. longisetus* (Thiébaud) released for biocontrol of *Cx. quinquefasciatus* in comparison with the existing copepod and fish populations found during our survey.

ACKNOWLEDGMENTS

We thank our Tropical Health Program colleagues, P. F. L. Boreham (QIMR) and J. H. Bryan (University of Queensland) for manuscript review, D. Battistutta and G. Williams for statistical assistance and J. W. Reid (Smithsonian Institution) for identifying the copepods used. We also acknowledge assistance given by other team members of the Federal University of Ceará and National Health Foundation, Fortaleza working within grant No. 403788/90.0 funded by CNPq, Brasilia and coordinated by A. W. Vasconcelos.

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