Am. J. Trop. Med. Hyg., 59(4), 1998, pp. 657–660

ERADICATION OF *AEDES AEGYPTI* FROM A VILLAGE IN VIETNAM, USING COPEPODS AND COMMUNITY PARTICIPATION

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Abstract. In northern Vietnam, copepods of the genus *Mesocyclops* were used for biological control of *Aedes aegypti*, the principal vector of dengue viruses, by inoculation into wells, large cement tanks, ceramic jars, and other domestic containers that served as *Ae. aegypti* breeding sites. The use of *Mesocyclops* was complemented by community participation with respect to recycling to eliminate unused and discarded containers that collected rainwater and provided *Ae. aegypti* breeding sites that could not be treated effectively with *Mesocyclops. Aedes aegypti* disappeared from 400 houses of the treated village in August 1994 and has not reappeared, a result of particular significance, because there are virtually no other recorded instances of eradicating this mosquito anywhere in the world during the past 25 years, and certainly not with community-based approaches. When used in combination with community recycling, *Mesocyclops* is an easy and inexpensive method of *Ae. aegypti* control that should be effective for many communities in Vietnam and elsewhere.

Dengue and dengue hemorrhagic fever (DHF) are mosquito-borne viral diseases known to occur in more than 100 countries, placing two-fifths of the world's population at risk.[1,2] Unpublished World Health Organization data indicate there are 30–60 million cases each year, including thousands of deaths from DHF (particularly among children). Dengue fever first appeared in Vietnam at Hanoi and Haiphong in 1959 and in South Vietnam in 1960. The Ministry of Health recorded 1,577,452 cases and 12,942 deaths from 1963 to 1994.[3]

In 1993, the World Health Assembly officially designated dengue control and prevention as a high priority, and in 1995, a global strategy was drafted that combined the use of new surveillance and control tools, intersectoral involvement, better training, and community participation.[2] However, success has been limited by a lack of effective methods to control the principal vector, *Aedes aegypti* (L.), an urban mosquito that breeds in water storage containers as well as discarded containers that collect rainwater. *Aedes aegypti* virtually disappeared from many countries 30 years ago, when DDT was widely used, but it became common again after DDT use diminished.[4] During recent years, urban expansion in Vietnam and other Third World countries has led to a proliferation of *Ae. aegypti* breeding sites. The lack of infrastructure, such as piped water, makes water storage containers a common necessity, and inadequate solid waste management has generated an accumulation of discarded containers.[5]

Herein we report the success of a relatively new control method: cyclopoid copepods. Although it has long been known that these tiny crustaceans prey upon first-instar mosquito larvae,[6] the unique potential of cyclopoids for mosquito control was only realized in 1981.[7] Predacious cyclopoids are particularly effective because of their broad diet, consisting of algae, protozoa, rotifers, and most aquatic animals up to their own size and because they do not depend on the supply of mosquito larvae.

Although cyclopoids offer possibilities for controlling many different kinds of mosquito larvae,[8–10] most of their use to date has been directed toward container-breeding *Aedes*. Various species of *Mesocyclops, Macrocyclops, Megacyclops*, and *Acanthocyclops* have been field tested in a variety of *Aedes*-breeding habitats in the Pacific, the Americas, and Asia, including wells, water storage containers (e.g., tanks, 200-liter drums, and ceramic jars), discarded tires, and land crab (*Cardisoma carnifex*) burrows.[11–14]

Local populations of *Ae. albopictus* (Skuse) have been eradicated by introducing cyclopoids into tires in New Orleans, Louisiana,[15] where *Mesocyclops* has become an operational part of municipal mosquito control.[10] In the tropics, where dengue is an ongoing problem, *Mesocyclops* have generally proved effective, reducing *Ae. aegypti* production by 95–100%. Total elimination of *Ae. aegypti* production from wells has been achieved with *Mesocyclops* during small-scale field trials in French Polynesia, Laos, and northern Australia.[16–18]

The critical factor for success with *Mesocyclops* has been their ability to survive in containers under conditions of everyday use. In some field trials to date, *Mesocyclops* have been lost from water storage containers when all the water is removed for use or containers are cleaned.[14] Although this difficulty is easily overcome if people restock their containers with *Mesocyclops*, large water storage containers in Vietnam are particularly suitable for *Mesocyclops* because they normally contain water and typically are cleaned about once a year.

As part of a World Health Organization consultancy in 1989, one of the authors (BHK) collected *Mesocyclops* in Hanoi[19] and with another author (VSN) initiated investigations of their efficacy with respect to control of larval *Ae. aegypti*. This paper describes the success achieved on a community scale from 1993 when a pilot project was commenced at Phanboi village, 31 km east of Hanoi, which resulted in *Ae. aegypti* eradication.

MATERIALS AND METHODS

Field site and strategy. After discussion with local leaders, Phanboi village (31 km east of Hanoi in the Disu commune, Myvan district, Haihung Province) was selected as the site for a pilot project. Nhanvinh village (1 km from Phanboi) was selected as the negative control. Both villages are composed of 400 houses. The kinds and numbers of *Ae. aegypti* breeding sites in the two villages were nearly identical, consisting mainly of outdoor cement tanks (average capacity = 2,700 liters) to store rainwater from roofs, ceramic jars (average capacity = 21 liters) to store water for immediate household use, and unused and discarded containers (e.g., cans, glass jars, metal parts, bottles, old tires, and broken bowls). Backyard wells were common. The two villages, as with all others in the district, had large numbers of *Ae. aegypti* and a history of dengue/DHF epidemics in 1983, 1987, and 1991.

In February 1993, project staff introduced 250 field-collected copepods into Phanboi's pond-like village well (20 m diameter). They also introduced 50–100 field-collected copepods into each of the 65 backyard wells, 352 cement tanks, 178 ceramic jars, and 119 miscellaneous unused or discarded containers observed to be in the village at that time. Copepods for the introductions were collected from Hoan Kiem Lake and Truc Bach Lake in Hanoi and from water storage tanks in Hai Hung town (60 km east of Hanoi) with a 100 μm mesh net. In June 1993, copepods from the same collection sites were introduced by project staff to all Phanboi containers that sampled negative for *Mesocyclops.*

The community was not asked to take an active role in the project during the first 15 months, but in March 1994 every household in the treatment village was given field-collected cyclopoids, and community involvement was initiated. The use of *Mesocyclops* for dengue control (including a live demonstration of *Mesocyclops* predation on *Ae. aegypti* larvae) was explained to village leaders, health workers, and representatives from the youth union and women's union, who conveyed the information to every household in the village. When cleaning containers, villagers were advised to retain some water with *Mesocyclops* in a bucket so that they could be returned to the container after cleaning.

At the same time, the village intensified a recycling program that provided an incentive to get rid of discarded and unused objects such as bottles, tin cans, tires, plastic bowls, and battery cases that could serve as breeding sites for *Ae. aegypti.* It was previously possible for mosquitoes to breed in these unused containers while villagers waited for collectors to buy them for recycling. After March 1994, the collections were frequent enough and complete enough to eliminate virtually all unused containers from the village.

Monitoring of copepods and mosquitoes. Monitoring began one month before the first copepod introduction and has continued until the present time. Thirty randomly selected houses in the treatment village and 30 houses in the control village (7.5%) were inspected monthly for the presence of cyclopoids and *Ae. aegypti* larvae in containers and adult *Ae. aegypti* inside the houses. The same houses were inspected each month. Resting adult *Ae. aegypti* were assessed by two persons using a flashlight and glass tube for 15-min collections per house. Surveys for the presence of cyclopoids and *Ae. aegypti* larvae in all village containers were conducted in May–June 1993, March 1994, May 1994, and October 1994. Nets of 100 μm mesh were used for tanks and jars whereas nets and funnel traps[20] were used for wells.

During November 1994 when all introductions were complete, samples of copepods were taken for specific identification. However, it should be noted that community translocation of copepods continued throughout the study period, on the basis of perceived need. In July–August and November 1996, additional surveys of 100 houses (25%) in each village were done to confirm the eradication.

RESULTS

Identification of copepods. The introductions consisted of *Mesocyclops woutersi* Van de Velde, *M. thermocyclopoides* Harada, and *M. ruttneri* Kiefer, often mixed with four species of *Thermocyclops* with some *Eucyclops* and *Microcyclops.* Voucher specimens of all species are lodged with the National Museum of Natural History, Smithsonian Institution (Washington, DC).

At the National Institute of Hygiene and Epidemiology Laboratory in Hanoi, cultures were established and predatory ability was established first in beakers containing 0.2 liters of water and later in aquaria containing 5–10 liters of water. The *Mesocyclops* spp. killed > 30 of newly emerged *Ae. aegypti* larvae per day whereas the other three genera were not effective predators (Nam VS, 1995, *Characteristics of Biology and Ecology and Measures for the Prevention and Control of DF/DHF in North Vietnam.* PhD thesis, University of Hanoi, Hanoi, Vietnam).

Presence of copepods in containers. The number of cement tanks that contained cyclopoids in the treatment village varied from 87% to 95% over the monitoring period. *Mesocyclops woutersi* was the only *Mesocyclops* species in tanks (68% of the tanks) when species identifications were done in November 1994. An additional 23% of the tanks contained either *Thermocyclops crassus* (Fischer) or *T. schmeili* (Poppe & Mrazek) at that time.

The number of ceramic jars with cyclopoids varied from 56% to 83% during the monitoring period. In November 1994, *M. woutersi* and *M. thermocyclopoides* were in 39% and 33%, respectively, and small numbers of *M. ruttneri* were in 11% of the jars, all of which also contained *M. woutersi* or *M. thermocyclopoides. Thermocyclops crassus,* which came from natural populations in village wells, was in 28% of the jars but always with *M. woutersi. Mesocyclops woutersi* was common in most wells.

Monitoring of larval and adult *Ae. aegypti.* The number of *Ae. aegypti* larvae/house in the treatment village was 30–97% less than the control village during the first 12 months after *Mesocyclops* introduction (Figure 1A). The number of larvae/house decreased to 87–99% less than that of the control village during the five months immediately after community involvement (including intensified recycling) began, and in August 1994 the larvae disappeared. No *Ae. aegypti* larvae have been observed in the treatment village since then.

The number of adults in the treatment village was 30–100% less than in the control village during the first 12 months after introduction of *Mesocyclops* (Figure 1B). Adults decreased to 87–99% less than the control village during the five months after recycling began, and then they disappeared. Except for a single individual in November 1994, no *Ae. aegypti* adults have been observed in the treatment village since then. At Phanboi in July–August and in November 1996, 225 and 286 containers, respectively, were inspected and no immature forms were found (Figure 1A). Similarly, no adults were detected (Figure 1B), representing an absence for two years.

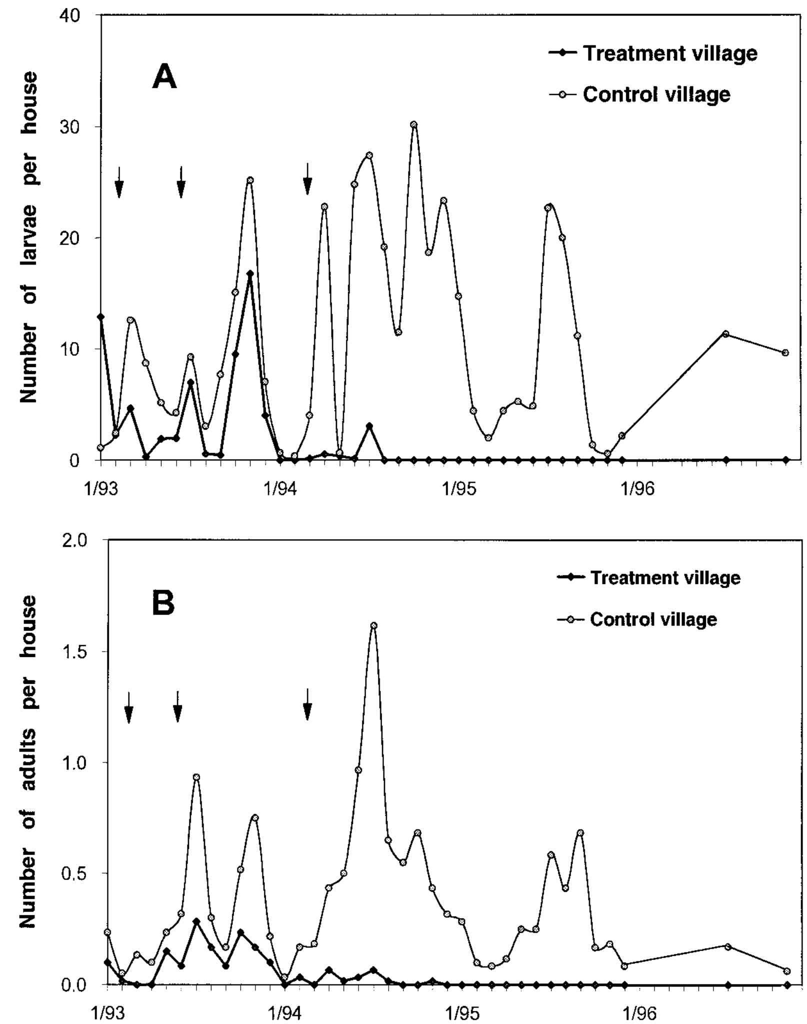

FIGURE 1. *Aedes aegypti* abundance in the treatment and control villages. **Vertical arrows** indicate times of *Mesocyclops* introduction to the treatment village. **A,** *Ae. aegypti* larvae; **B,** *Ae. aegypti* adults.

DISCUSSION

With the possible exception of Cuba,[21] the disappearance of *Ae. aegypti* from Phanboi is the only recorded eradication of *Ae. aegypti* anywhere in the world during the past 25 years. Why was *Mesocyclops* so successful in Phanboi? First, the way that people in Phanboi used their water storage containers made it possible to keep copepods in most of the containers with relatively little effort. Community involvement and recycling, which eliminated containers that could not be treated with *Mesocyclops*, were also important because *Ae. aegypti* was eradicated only after these components were part of the project. We have done numerous surveys for *Ae. aegypti* in northern Vietnam where recycling occurs as an economic activity. Other villages in Di Su commune, at Lac Vien commune in Haiphong, and at Nghia Dong in Nam-ha Province are always positive for *Ae. aegypti* in the absence of *Mesocyclops.*

Aedes aegypti disappeared from Phanboi village although *Mesocyclops* apparently were not in all of the water storage containers. Part of the explanation lies in the egg sink effect of *Mesocyclops*. Every container with *Mesocyclops* was not only prevented from producing *Ae. aegypti* but also functioned as a sink for *Ae. aegypti* eggs by competing for oviposition with containers that did not have *Mesocyclops.* Because *Mesocyclops* are only 1 mm long and may have been at low densities in some containers, undersampling is also likely. Furthermore, it is likely that other sources of mortality, e.g., mosquito coils and spray cans of insecticide plus with jars, regular emptying for domestic consumption, may have contributed.

Combining *Mesocyclops* in water storage containers with community cleanup of other containers (for economic gain) appears to be very promising for *Ae. aegypti* control in villages such as Phanboi. If *Mesocyclops* proves as successful in other villages, it should merit widespread application in Vietnam and possibly other parts of Southeast Asia. The use of *Mesocyclops* does not demand excessive time, effort, or technical expertise from the community, and the cost of providing local natural populations of *Mesocyclops* is extremely low. It is only necessary to provide a small number of *Mesocyclops* for introduction into key village sites (i.e., tanks, wells) because numbers increase quickly.

The effectiveness of *Mesocyclops* can best be ensured by getting them into as many containers as possible. This can be facilitated by ensuring that all wells are stocked so *Mesocyclops* are continually reintroduced to containers such as ceramic jars when they are used to store well water for household use. Avoiding the introduction of non-larvivorous copepods should minimize competitive exclusion of *Mesocyclops* by other copepods. Mass production of *Mesocyclops* in the laboratory is simple and inexpensive and can provide millions of *Mesocyclops* if suitable natural sources are not available.[22]

Acknowledgments: We thank Dr. Hoang Thuy Nguyen (former Director, National Institute of Hygiene and Epidemiology, Hanoi), Dr. Tran Van Tien (Deputy Director, National Institute of Hygiene and Epidemiology), and Dr. L. Self (World Health Organization, Manila) for advice and support of this project. The field staff were Phan vu Diem Hang, Tran Vu Phong, Nguyen Tu Bin, Bui Huy Nhanh, and Luong Duc Dan. Maria Holynski collaborated in identifying *Mesocyclops* species. Mieu Nguyen and other staff of the New Orleans Mosquito Control Board provided laboratory work in support of this study.

Financial support: This project was funded by the World Health Organization, Western Pacific Region. Financial support for cyclopoid species identifications was provided by The Rockefeller Foundation (Health Sciences Division) to the Department of Tropical Medicine, Tulane University (New Orleans, LA).

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