EFFECT OF RICE HUSBANDRY ON MOSQUITO BREEDING AT MWEA RICE IRRIGATION SCHEME WITH REFERENCE TO BIOCONTROL STRATEGIES

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ABSTRACT. A study was carried out at Mwea Rice Irrigation Scheme, Kenya, to assess the impact of rice husbandry on mosquito breeding and identify indigenous biocontrol agents with potential for controlling mosquito breeding in the scheme. The study established a close relationship between the schedule of the farming practices (particularly the flooding phase) and mosquito breeding. Two groups of agents, entomopathogenic bacteria (*Bacillus thuringiensis* var. *israelensis*) and larvivorous fish, were identified. Laboratory evaluation of the agents produced encouraging results. The bacterial isolates showed broad-spectrum larvicidal potency against *Anopheles, Culex* and *Aedes* mosquito larvae and 2 of the fish species, *Tilapia zilli* and *Oreochromis niloticus,* demonstrated a strong predation for a mosquito larval diet. To facilitate their use in effective biocontrol strategies, the agents would require further evaluation under field conditions.

INTRODUCTION

The intimate relationship between the schedule of rice husbandry and mosquito breeding has long been recognized in several countries (Surtees 1970, Grainger 1947, Russell and Rao 1940). In their study on mosquitoes of the Kano Plain (Kenya), Chandler et al. (1975) indicated that in the rice field environments, numbers of the main malaria vectors were higher than in non-irrigated areas. Sen (1948) attributed the breeding patterns of *Anopheles* spp. to rice cultivation practices. In Japan, Makiya (1967) related breeding densities of *Culex tritaeniorhynchus* Giles to increase in rice height, and Reuben (1971) observed a similar pattern in the breeding of culicines in rice fields of Madras. In Californian rice fields, Womeldorf and Whitesell (1972) demonstrated the relationship between the breeding of *Culex tarsalis* Coq. and *Anopheles freeborni* Aitken and water depth and height of the rice.

Biocontrol agents are becoming increasingly popular in controlling larval mosquito populations. Among these natural agents, bacteria and larvivorous fish are the 2 main groups that are widely used in controlling breeding in rice-field environments. Varieties of *Bacillus thuringiensis* Berliner and *Bacillus sphaericus* Neide have been widely tested and employed for larval mosquito control in various situations and have proved effective in rice fields (Hembree et al. 1980, Lacey et al. 1986, Bowles et al. 1990). As biological mosquito-control agents, larvivorous fish such as *Gambusia* and *Tilapia* have also been remarkably successful, and they are particularly effective for controlling mosquito breeding in rice-field situations (Castleberry and Cech 1990, Hoy and Reed 1971, Hoy et al. 1971, Gerberich and Laird 1968, Bay 1967).

This study was carried out with the two-fold objective of monitoring the seasonal changes in mosquito larval populations at Mwea Rice Irrigation Scheme in relation to the cultivation cycle to determine the impact of the farming practices on mosquito breeding, and of identifying potential biocontrol agents for controlling mosquito breeding in the scheme.

MATERIALS AND METHODS

Study area: Covering an area of 12,000 ha, the Mwea Rice Irrigation Scheme is located approximately 100 km NE of Nairobi (Kenya) on the foothills of Mount Kenya, at an elevation of 1,159 m above sea level and latitude 0° 4′ south. The irrigation water is tapped from the Nyamindi and Thiba rivers and fed to the scheme by gravity. A population of 50,000 people in 36 villages is supported by the scheme. The annual rainfall varies from a maximum of 1,626 mm to a minimum of 356 mm, averaging 950 mm/year. Maximum precipitation takes place in April/May (long rains) and October/November (short rains). The average temperatures are in the range of 16.0–26.5°C and the relative humidity varies from 52 to 67%.

Rice cultivation cycle: Only one crop of rice is grown annually. Although the constituent farming activities do overlap within the annual cycle, the schedule of husbandry could be conveniently described under 4 operations: land preparation, nursery development, transplantation and harvesting. The cycle begins with preparation of the land, which involves burning of vegetable waste, dredging and repairs of canals, roads and drains. Land preparation occupies approxi-

mately 3 months (December–February) before the long rainy season begins in March. During the long rains (March–May), there is widespread natural flooding but no rice is planted. Cultivation resumes shortly after the rainy season and culminates in ploughing and flooding through the irrigation system. Following ploughing, seed rice is sown in the nursery beds situated in the corners of the paddies. Within 4–5 wk (July/August), the young seedlings are transplanted onto the main paddies. Some 5–6 wk after transplantation, chemical insecticides (Sumithion® and Carbofuran®) are applied to control rice pests, mainly stem borers and leaf miners. The rice is mature by the end of November when the paddies are drained to facilitate harvesting.

Larval sampling: By means of a 500 ml dipper, mosquito larvae were collected weekly (4 times per month) throughout a 1-year period (January–December 1991). Collections were made from 2 sites each of 4 habitats: rice paddy, irrigation canal, pool and pond. Pool and pond were included among the sampling habitats so that larval production could be compared in relation to irrigation-related systems (rice paddy and irrigation canals) and nonirrigation-related habitats (pond and pool). It is relevant that some pools were created from runoffs of flooded drains and irrigation canals and could therefore be regarded as irrigation-related. The larvae were counted and grouped into their respective *Anopheles* and *Culex* species. On each occasion, 20 dips were made at 2 sites of each habitat and the number of larvae counted in each dip were pooled together to represent the total larval collection at the habitat.

Biocontrol agents: In two separate publications (Asimeng and Mutinga 1991, 1992a), the authors have given comprehensive accounts of the biocontrol agents (mosquito-toxic bacteria and larvivorous fish) identified in the study area. The source materials (mosquito larvae, soil, mud) for isolation of bacteria were collected from mosquito breeding habitats. Several varieties of bacteria were isolated using a medium containing yeast extract, tryptone, and NaCl (Miller 1972). The isolates were tested for their larvicidal property and the promising materials were preserved for further evaluation in powder formulations prepared by the lactose coprecipitation method (Dulmage et al. 1970). Samples of fish were collected from irrigation canals and ponds, and transported to the laboratory for identification and evaluation. Affinity for larval diet was evaluated using *Tilapia zilli* and *Oreochromis niloticus* on *Aedes aegypti* (Linn.). *Tilapia zilli* has been evaluated in the field to assess its efficiency under natural conditions (Asimeng and Mutinga 1992b).

Data analysis: Monthly larval collection and rainfall data supplied by Mwea Irrigation Agromet Station were subjected to Pearson's correlation analysis after log transformation of the larval densities.

RESULTS

Although a significant number of larvae were collected in April during the peak of the long rainy season, in general, the abundance of larvae was negatively correlated with rainfall (Fig. 1); r was not significant at $P = 0.05$ ($r = -0.005$, $P > 0.98$). A total of 29,763 larvae were collected; the monthly mean numbers at the 4 habitats are shown in Fig. 2.

The irrigation system comprising the canal network, drains and paddies was flooded for 5 months (July–November). During this period, a total of 18,689 larvae were collected; this number represented 63% of the entire collection (29,763) for the year. Larvae were collected from the rice paddies, 11,375 (60.9%); pools, 7,277 (38.9%) and ponds, 37 (0.2%). No larvae were collected from the fast-flowing irrigation canals. Samples of *Anopheles* larvae were taken in larger numbers than those of *Culex* species, particularly in the paddies, a few weeks after transplantation. The peak of larval density was recorded in August when 7,143 larvae (11 larvae/dip) were collected; most of those (4,965 larvae) were collected from the rice paddies (8 larvae/dip).

In the dry season (December–March), water in the canal network was dried up in most places. March is included in the dry season as the greater part of it was dry except toward the end of the month when a significant amount of rainfall was recorded. The drying process resulted in creation of a series of isolated pools all along the canal that provided the major habitat for mosquito breeding during this period. In the dry season, 1,448 larvae were collected, accounting for 5% of the annual total; 1,438 (99.3%) samples were collected from the canal pools and 10 (0.7%) from the ponds. Larvae of *Anopheles* were represented in smaller numbers than those of *Culex* species, which thrived better in the canal pools littered mostly with decomposing vegetable matter of rice origin (Fig. 3).

During the long rains (April–June), samples of larvae were collected from all the habitats except the unflooded rice paddies. A total of 9,626 larvae (32% of the annual total collection) were collected during this season; of these, 9,413 (98%), 109 (1%) and 104 (1%) originated from pools, irrigation canals and ponds, respectively.

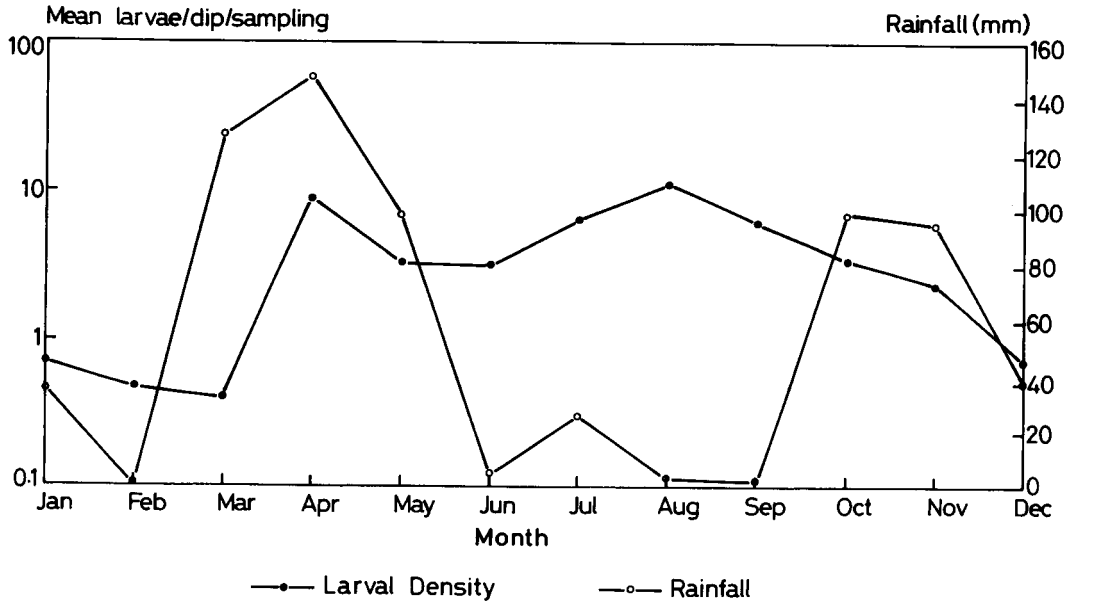

Fig. 1. Monthly larval density in relation to rainfall from January through December 1991.

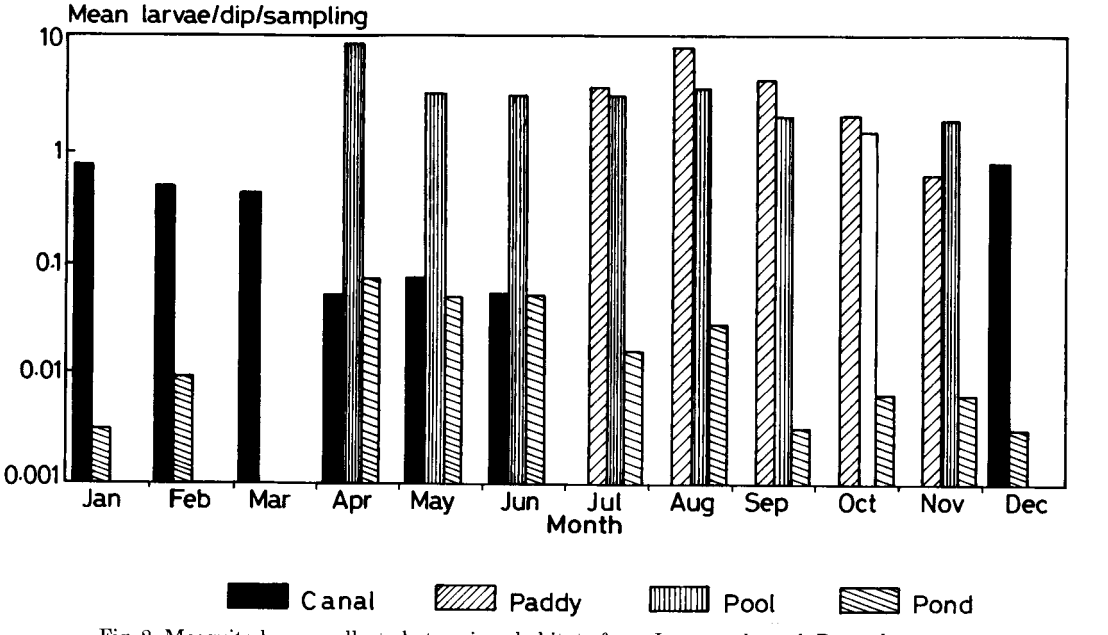

Fig. 2. Mosquito larvae collected at various habitats from January through December 1991.

Anopheles larvae were collected mostly during the flooding phase, particularly during the post-transplantation period in August (Fig. 3). The major *Anopheles* larvae were *An. arabiensis* Patton and *An. pharoensis* Theobald which constituted approximately 84 and 15%, respectively. Larvae of *An. funestus* Giles, *An. pretoriensis* Theobald and *An. maculipalpis* Giles formed the minor *Anopheles* species.

Laboratory evaluation of the feeding habits of *T. zilli* and *O. niloticus* suggested that both species were larvivorous and showed a marked interest in mosquito larvae (Asimeng and Mutinga 1991). Fingerlings of *Oreochromis niloticus* and

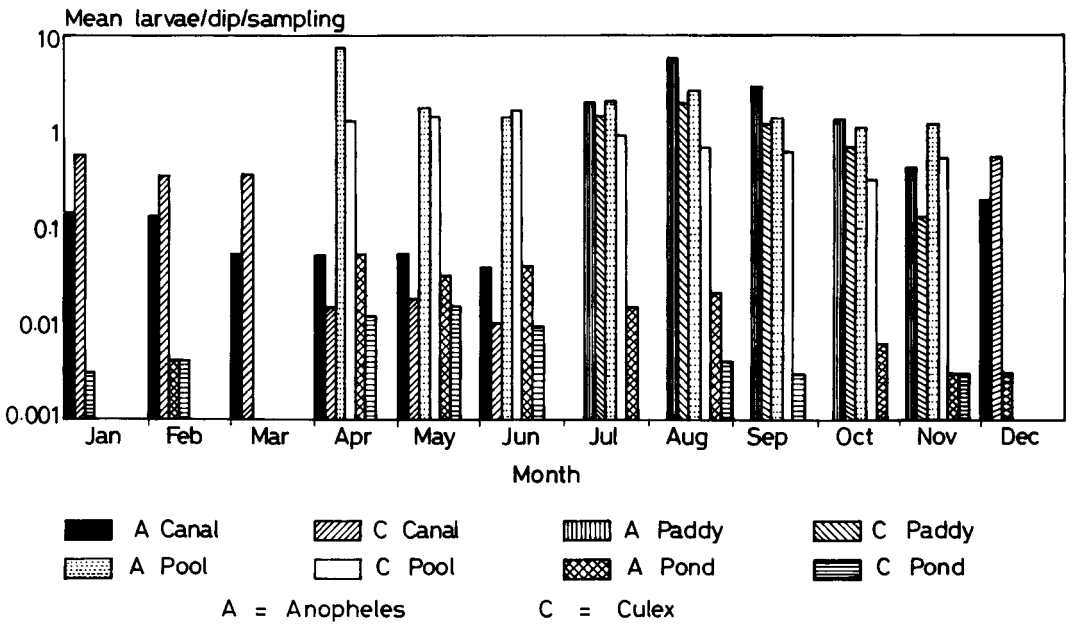

Fig. 3. Seasonality of *Anopheles* and *Culex* larvae at various habitats.

T. zilli were used in feeding experiments. To facilitate adequate acclimatization in the laboratory, the fish were held in glass tanks containing plankton-laden pond water maintained at 22°C for at least 8 wk prior to the start of the feeding experiment. They were distributed 8 to a tank measuring 60 × 30 × 30 cm. Their planktonic diet was supplemented by crushed commercial pond pellets containing proteins, vitamins, minerals and essential amino acids, rationed on a daily basis at 250 mg per fish. The feeding experiment was performed on one fish at a time, using one specimen just once. A fish was removed from the holding tank and transferred into an experimental tank containing pond water. It was held in the experimental tank for 1 h before the start of larval feeding. Following the 1 h period, 500 third stage larvae of *Aedes aegypti* were introduced into the experimental tank. After a period of 15 min, the fish was removed and the number of larvae eaten was estimated. The experiment was repeated 4 times over a range of temperature (20, 25, 30 and 35°C) and at different pH values (7.0, 7.5, 8.5 and 9.5). An average of 300 larvae were eaten by one fish in 15 min under optimum conditions of temperature (25°C) and pH (7.0); *T. zilli* consumed more larvae than *O. niloticus*.

Several samples of entomopathogenic bacteria that have demonstrated toxicity against *Anopheles, Culex* and *Aedes* mosquito larvae were isolated (Asimeng and Mutinga 1992a). All the isolates (B42, B51, B53, B54 and B55) have now been identified and registered as *B. thuringiensis* var. *israelensis* at the Pasteur Institute in Paris.

DISCUSSION

It has been demonstrated in the present study that the prevailing rainfall pattern at Mwea Rice Irrigation Scheme generally has insignificant bearing on mosquito breeding, which is rather closely associated with the schedule of rice husbandry with particular reference to the flooding phase. The most critical period in the cropping cycle with respect to mosquito breeding occurred during the flooding season (July–November), when the entire irrigation system was inundated with water. The flooded paddies provided a suitable environment that sustained prolonged and prolific breeding. The period of transplanting the young seedlings onto the main paddies was of particular interest. Numerous shallow and sunlit pools and puddles, resulting from the footprints of the workforce, created extensive habitats conducive for intensive breeding, particularly of *Anopheles* species. This might have accounted for the overwhelming number of larvae collected in August after the period of transplantation. Pools formed from runoffs of overflowing canals and drains equally generated widespread potential breeding sites. During the dry season, breeding was largely confined to the isolated

canal pools occurring especially along the minor canal network.

The application of chemical insecticides against rice pests could have detrimental effects on the bionomics of the complex rice field environment. In addition to the target rice pests, the chemical pesticide could kill both mosquito larvae and their potential predators, such as adult Coleoptera and Diptera, and other mosquito larvae and frogs, together with large numbers of nonpredacious insects, which provide alternative prey for predators. Because the predator and nonpredacious populations take longer to recover from the effects of the chemical insecticide than do the mosquitoes, an exceptional outburst of mosquitoes occurs a short time after the pesticide application, as observed in the present study. As reported by Gillies and de Meillon (1968), *Anopheles gambiae* Giles has an extremely rapid life cycle under local conditions (egg to adult, 6–9 days).

The biocontrol agents identified and isolated in this study could be used on their own or incorporated in an integrated program to control mosquito breeding in the irrigation scheme. Fish have great potential as biocontrol agents against larvae and pupae of mosquitoes. The relatively small numbers of mosquito larvae collected from the pond habitat could be attributable to effective predation of the larvae by larvivorous fish, which were observed in large numbers in this biotope. For control strategies, the identified larvivorous fish could be collected and raised in sufficient numbers in man-made ponds to facilitate stocking of the rice fields at a strategic time shortly after the transplantation period. Controlling breeding in pools and drains could be effectively carried out using the mosquito-toxic bacterial isolates in appropriate formulations. Such preparations could also be used to supplement control programs in other breeding sites.

To facilitate an effective biocontrol program using the biocontrol agents identified in this study, their potential in controlling mosquito breeding should be evaluated under local field conditions to determine possible environmental factors likely to influence their efficiency in nature. Furthermore, assessment of the effects of the rice pesticides currently in use on aquatic life would be beneficial. If necessary, a suitable microbial insecticide for rice pest control would serve as a satisfactory substitute.

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