# ARTICLES

## BIOLOGICAL MOSQUITO CONTROL FURTHERED BY ADVANCES IN TECHNOLOGY AND RESEARCH

E. F. LEGNER[1] AND R. D. SJOGREN[2]

ABSTRACT. Natural enemy systems that persist in the environment increase their effectiveness through field reproduction, and may coevolve with their hosts to minimize the development of resistance to pesticides. Some biotic insecticides are commercially available for short-term direct mosquito control, which may be vital to exploit for malaria control in countries where technology is not advanced. A few examples of long-term persistent biological controls exist. A comprehensive view of biological control should differentiate geographic areas, habitat characteristics and levels of human tolerance, and distinguish purely mosquito annoyance problems from life-and-death disease vector impacts on humans in technologically underdeveloped areas. The future of biological mosquito control is dependent on additional research, especially the discovery of capable new species and strains of organisms. With resistance to insecticides a compounding problem, biological control in concert with practical breeding site modification, may be the only long range tactic to reduce mosquito densities over a broad expanse of the world.

### INTRODUCTION

A widely accepted definition of biological control from a medical viewpoint recently stated in the "Sixth Report of the World Health Organization Expert Committee on Vector Biology and Control" (1982) is, "The control of pests, including the vectors of human disease, by the direct or indirect use of natural control agents with or without their metabolites." Implied in this definition is the presence of some living natural biotic agent either in the active stage or at some time prior to field application, such as the toxin in *Bacillus thuringiensis* subsp. *israelensis* de Barjac, generally referred to as *B. thuringiensis* H-14. The first effective effort to organize information on biological control of medically important arthropods was made by Jenkins (1964) at the invitation of the World Health Organization, and Dr. Marshall Laird. Following accelerated research emphasis in the 1960's and 1970's, new reviews have dealt with various aspects of mosquito biological control (Bay 1973, Bay et al. 1976, Chapman 1974, Garcia and Dahlsten 1980, Hertlein et al. 1980, Laird 1971a, 1971b, 1977, 1980; Legner et al. 1974, National Academy of Sciences 1973, Petersen 1973, Platzer 1981, Roberts and Strand 1977, Roberts and Castillo 1980, Roberts et al. 1983; Service 1983, World Health Organization 1973, 1982).

Although research has shown that entomopathogens and predators can effectively reduce mosquito numbers, the practical value of biological control was questioned by Service (1983). As is typical of biological control, reductions of mosquito larvae and pupae are usually not immediate as is customarily experienced with insecticides. Although occasionally mosquito reductions at differing levels have been continuous, in very few cases have they persisted over long periods of time (Hauser et al. 1976; Legner 1983, Legner and Fisher 1980, Legner and Murray 1981, Legner and Pelsue 1983, Petersen et al. 1978b). Field studies of some semi-permanent habitats also have shown the value of resident natural enemies in maintaining the mosquito larval densities at levels which do not require cultural or insecticidal control measures (R. D. Sjogren, unpublished data, Legner et al. 1975b, Walters and Legner 1980). Preservation of the natural predator complexes does in some cases require deliberate management and a continuous awareness of their importance, which naturally restricts their maximization in these instances to technologically advanced countries. Recognition of the existence of effective predator complexes can greatly reduce the cost of mosquito abatement, but usually requires intensive studies over several years and seasons (Case and Washino 1979, Collins and Washino 1978, Glenn and Chapman 1978, Hauser et al. 1976, 1977; Washino 1981). Where field reproduction of natural enemy complexes occurs, the possibility of coevolution with their mosquito hosts exists, eliminating host resistance, and resulting in greater predator/prey stability at lower mosquito larval population densities.

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[1] Entomologist and Professor, Division of Biological Control, Department of Entomology, University of California, Riverside, CA 92521.

[2] Director, Metropolitan Mosquito Control District, 2380 Wycliff Street, St. Paul, MN 55114.

Sometimes periodic addition of natural enemies to chronic mosquito breeding areas is effective in reducing breeding to acceptable levels. Various organisms have given encouraging results in field trials, even under environmentally polluted situations. Included are turbellarians, fungi, some bacteria, coelenterates, and the predatory mosquito genus *Toxorhynchites*. Although some species do not possess all the attributes of a good biological control organism (Service 1983), there are certain habitats in which they may afford the only practical control of any kind. (Ali and Mulla 1983, Axtell et al. 1982, Chapman 1974, Des Rochers and Garcia 1984, Fochs et al. 1979, Garcia and Des Rochers 1979, Garcia et al. 1980, George et al. 1983, Gerberg and Visser 1978, Goldberg and Margalit 1977, Jaronski and Axtell 1982, 1983a, 1983b; Legner 1977, 1978a, 1979; Legner and Yu 1975, Legner et al. 1975c, Levy and Miller 1977a, 1977b, 1978; Merriam and Axtell 1982a, 1982b, 1983; Miura et al. 1980, Mulla et al. 1984, Palchick 1984, Schaefer and Kirnowardoyo 1983, Sjogren and Legner 1974, Sweeney et al. 1983, Yu and Legner 1976, Yu et al. 1974a, 1974b, 1975). However, to date mass production techniques are limited, and few species have been implemented practically (Jaronski et al. 1983, Laird and Miles, in press; Legner 1978b, Legner and Medved 1974[b], Legner and Tsai 1978, Legner et al. 1976, Lenhoff and Brown 1970, Medved and Legner 1974, Merriam and Axtell 1982b, Nelson 1979, Petersen and Willis 1972, Petersen et al. 1978a, Tsai and Legner 1977). Only *Romanomermis culicivorax* Ross and Smith, *Bacillus thuringiensis* H-14 and *Bacillus sphaericus* Neide are either presently available or could be shortly, for widespread use (Schaefer and Kirnowardoyo 1983, Petersen and Cupello 1981, Service 1983). When recycling of some of these organisms does occur, it is usually significant only during the year of application, with winter carryover not being high enough to evoke satisfactory mosquito reductions during the early part of the season of the following year.

Natural enemies may occasionally be added to mosquito breeding habitats where they persist through the mosquito season, and multiply over time. The minnows, *Gambusia affinis* (Baird and Girard), and *Poecilia reticulata* Peters are most commonly used in this manner (Bay 1972, Bay et al. 1976, Gall et al. 1980, Gerberich and Laird 1968, Hoy and Reed 1970, 1971; Hoy et al. 1971, 1972; Mulligan et al. 1983, Sasa and Kurihara 1981). The use of native fish also appears promising (Legner and Medved 1974a, Legner et al. 1975a, Walters and Legner 1980). Three species of cichlids, *Sarotherodon (Tilapia) mossambica* (Peters), *S. hornorum* Trewazas and *Tilapia zillii* (Gervais), have become permanently established in ca. 2,000 ha. of *Culex tarsalis* Coquillett breeding habitat in the irrigation system of southeastern California. In these areas, mosquitoes are controlled by the dual action of the fish feeding on protective aquatic vegetation, reducing favorable mosquito breeding habitats, and by direct predation of mosquito eggs, larvae and pupae (Legner 1978a, 1983; Legner and Fisher 1980, Legner and Medved 1973a, 1973b; Legner and Murray 1981, Legner and Pelsue 1983, Legner et al. 1975d). This unique example of persistent biological mosquito control is dependent on favorable water temperatures and a continuous supply of irrigation water to canals and drainage ditches that allowed the establishment of the subtropical cichlids. It is probably applicable only to areas where similar sophisticated water management can guarantee a permanent water supply.

Another case of persistent biological mosquito control is in the paved and unpaved river drainages of southwestern California where aquatic vegetation can accumulate around debris (i.e., boards, tires and other trash), providing protective niches for *Cx. tarsalis*. Two species of cichlid fish, *S. hornorum* and *S. mossambica*, have become established at high population densities, and constantly forage at these sites (Legner and Pelsue 1980, 1983), eliminating both the aquatic vegetation and the immature mosquitoes. However, the principal food to sustain these fish is chironomid larvae, which allows them to build up annually to the large numbers necessary for effective aquatic weed control (Legner and Medved 1973b, Legner et al. 1980). Persistence of these subtropical species in winter is apparently dependent on an artificial source of warm water supplied by a power generating plant adjacent to one of the paved channels.

## PRACTICAL CONSIDERATIONS

The relative stability of aquatic habitat is a principal determinant in the successful utilization of biological control agents for mosquitoes. To achieve predator-prey equilibrium at a level which will reduce or prevent disease transmission and/or maintain mosquito pest populations below the levels of human tolerance, cost considerations usually dictate that the control agent must reproduce after inoculation to achieve satisfactory control. For this reason, the genera *Culex*, *Culiseta* and *Anopheles*, which usually breed in permanent and semi-permanent water, have received more attention in biological control than have *Aedes* or *Psorophora*. In the sporadic, intermittent water habitat of the latter two genera, brood development can occur within a few days, requiring costly inundative releases of predators, parasites, or applications of mosquito pathogens which serve as biological insecticides. The highest levels of control have been achieved under low to moderate larval population densities. Massive synchronous brood development by *Aedes* or *Psorophora* may at times exceed the controlling capacity of naturally occurring predator complexes and/or economically feasible inoculation levels. In such cases, concurrent inoculations of other substances such as juvenile hormones and *Bacillus thuringiensis* H-14, can be used.

In stable aquatic habitats, *Gambusia affinis* continues to be the primary biological control agent used in early season inoculative releases. Adverse side effects of this species are the possible reduction of populations of economically important fish, and threatening of endangered species (Walters and Legner 1980).

However, there are no reports of this species causing phytoplanktonic blooms outside of the aquarium environment (Hurlbert 1975, Hurlbert and Mulla 1981, Hurlbert et al. 1972, Walters and Legner 1980). A number of effective insect and other invertebrate biological control agents of mosquitoes are also known (Chapman 1976). The most promising agents in permanent water habitat appear to be *Hydra,* freshwater flatworms and nematodes for which mass culture procedures have been devised, albeit they require further development. For sporadic floodwater habitats the most promising and potentially economical biological insecticide is the bacterial pathogen *Bacillus thuringiensis* H-14. However, due to the absence of residual activity in temporary water habitats, control economics imply that chemical insecticides will continue to be used in many of these areas due to cost considerations.

Environmental alterations to enhance the success of biological control agents are difficult, judging from empirical observations which indicate the need for water and nutrient level manipulations under some conditions to achieve a balanced invertebrate fauna to support necessary biological control agents, in the absence of mosquito larvae. Except where larval habitat elimination is practical, as with *Aedes aegypti* (Linn.) which breeds in man-made container breeding places, environmental manipulation will probably continue to play a secondary role to that of recognition of habitats in which conditions favor the maintenance of effective populations of natural enemies, due to the absence of specific data on site modification methods and their environmental impact. Economic considerations and conflicting land use patterns are factors limiting the number of aquatic habitats amenable to achieving dependable biological control.

## SPECIFIC BIOLOGICAL AND OPERATIONAL CONSIDERATIONS

A biological mosquito control agent must be dependable over a wide range of environmental conditions, particularly when operations personnel are required to recognize or predict when and where an agent will work effectively. Unpredictability of control is a frustrating and difficult complicating factor for field personnel, and one which control administrators strive to reduce as much as possible. A high level of training and time (i.e., monitoring) is essential for field personnel utilizing biological control agents to accurately recognize the varied environmental conditions including seasonal shifts in water quality, temperature and water permanence, which may result in control failure. To this end, there is a need for operational districts to support funding of the necessary studies. A high degree of confidence is also necessary in the introduction of a mass cultured organism (or any control), to insure with reasonable certainty that known levels of control will be achieved for the period necessary.

Logistically, if large tracts of land are contiguous and permit ready access for treatment and evaluation, a control agency finds greater possibilities to utilize a biological control agent. If the agent can survive and exert control only in select and dispersed locations, its use will be economically impractical.

The time spent in travel to a site to determine its suitability for certain biological control agents, then return for treatment and subsequent follow-up is often too time consuming for many small sites. Conventional methods of sweeping through an area, controlling all mosquito breeding in sites with insecticides or oils at scheduled intervals, is a more cost-effective approach, but speeds development of resistance problems. Implementing a number of different kinds of control methods within an area at the same time, each with its specific requirements, is more complex to supervise. Intermittent water breeding sites (i.e., where the site floods, dries and refloods during the year) hold water at erratic intervals, which frustrates the utilization of most biological control organisms, with the possible exception of parasitic mermithid nematodes (Petersen et al. 1978a, 1978b). Conscientious effort put forth is voided when erratic weather or human activities alter site stability. Reflooding results in mosquito return but little if any predator carryover.

A biological control program must have the total support of the program administrator to enable it to reach its highest level of effectiveness, including employees who frequently are skeptical and resist new technology. Combined with the natural reluctance of governing boards to appropriate funds for anything but traditional technology leads to a high risk venture for an administrator to support a large mass culture program for areas less than ca. 4,000 ha. of stable, semi-permanent water. For large habitats, such as found in rice growing areas of northern California costs benefits may be more favorable.

With operating cost efficiency and maximum possible control with available funding in mind, managers look closely at the cost per hectare per year required to achieve effective control, choosing the cheapest environmentally compatible control method possible. If a biological control agent is most feasible for large land tracts, a manager is not likely to put forth the effort necessary to use the control technique in small sites, particularly if the cost of training employees and repeated visits to the sites to determine if control is still in effect are high, and there is much likelihood that less than a dependable 95% overall control will be achieved.

Thus, a primary obstacle to a wider application of biological mosquito control in technologically advanced countries appears to be due to an inability of current biological control methods to achieve dependable results under the wide range of environmental conditions facing abatement agencies. The laboratory production, storage and distribution of biological control organisms requires a high degree of sophistication, which is usually unavailable at the operations level for most control agencies. Meanwhile, there are few commercial sources for most organisms in the numbers and at the costs required for mosquito control. A major thrust to an expanded reliance on biological controls will develop as new data are available on their effective management and chemical controls become less effective or more expensive.

## FUTURE CONSIDERATIONS

Biological mosquito control must be considered according to geographic area and habitat; and should always be integrated with other methods (Axtell 1979, 1983). There must be a recognition of those systems in which biological control is effective (e.g., irrigation system in the lower Colorado desert, waterfowl refuges, rice fields, etc.). More intensive studies of those systems are needed to learn why biological control works, so that correct decisions can be made for other areas, and certainly to preclude its attempted use where it is not possible (e.g., subtropical cichlids in colder areas or in semi-permanent water). New insights are also needed into what acceptable mosquito production levels are relative to breeding site locations and adjacent human population, which would allow less than 100% control with biologicals to be operationally feasible. A 12,800 sq-km. computerized regional mosquito management model is under development in the Minneapolis-St. Paul, Minnesota area to guide the control program, and in part allow greater use of combined natural mortality factors. The current goal of complete control for all breeding sites which drives most mosquito control programs in the United States, needs to be re-evaluated through development of population models. This would permit the weighing of adult emergence, distance from breeding sites, adjacent production terrain, wind direction, human population levels, daily survivorship rates, biting behavior, etc. for treatment threshold decisions.

An intensive study of certain invertebrates, especially flatworms, is needed. The wider appraisal and application of *Dugesia tigrina* Girard, *D. dorotocephala* (Woodworth) and the genus *Mesostoma* may be expected to produce worthwhile results for use in North America, based on the results of field experiments (Case and Washino 1979; George et al. 1983; Legner 1977, 1979; Yu and Legner 1976); and the discovery of or selection for new strains with resistance to environmental contaminants and insecticides may be possible. Flatworms of the genus *Mesostoma* are especially potent mosquito destroyers, and mass production of at least one species is needed to test inoculation effectiveness. Various species of freshwater hydra, fungi in the genera *Lagenidium* and *Culicinomyces*, mermithid nematodes, and predatory Hemiptera in the genera *Notonecta, Buenoa* and *Plea*, are promising candidates because they have mass production potential.

There is also a need to examine and categorize those ecosystems in which biological control effectiveness appears limited. Such areas include the tundra, wilderness snow melt, wilderness swamps, tree holes, artificial containers, and most pastures and intermittent rainwater depressions. It may be conceded, however, that this apparent limitation is due to a lack of research in such areas and that certain biological control possibilities do exist. The present inadequate technological status of many countries also precludes the effective use of most biological mosquito controls. Therefore, it has been suggested by Dr. Marshall Laird (personal communication) that our main thrust now in "third world" countries should not be for more research but for the speediest possible achievement of specific integrated control methodologies. This should include a combination of residual spraying, such as carbamates and other forms of adulticiding, larvicides, especially *Bacillus thuringiensis* H-14 and "third generation" pesticides such as the IGR/juvenile hormone mimic methoprene. This should be done against the background of adequate prior knowledge of the target mosquito and continuing source reduction with the fullest possible community participation.

As further research provides a fuller understanding of the factors associated with different resulting levels of control that can be achieved with biological agents, it will increase confidence in the predictability of their use and enhance their cost-effective integration into operational mosquito control programs. The question is not if biological mosquito control has a future (Service 1983), but whether that future can be enhanced through the proper research emphasis and technological development.

## ACKNOWLEDGMENTS

We express our appreciation to the following for their guidance and assistance in preparing this review: E.C. Bay, T. J. Case, R. Garcia, J. A. George, J. B. Hoy, M. Laird, R. Levy, F. R. S. Nelson, J. J. Petersen, R. C. Russell, C. H. Schaefer and R. A. Ward.

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## A NEW INTERNATIONAL AWARD

The Council for the International Congresses of Entomology recently approved a new "Distinguished International Award in Morphology and Embryology," to be presented to an outstanding morphologist or embryologist at each future Congress of Entomology.

The award was announced in the opening plenary session of the 17th International Congress of Entomology of Hamburg on August 20, 1984, by the Chairman of the Council, Dr. Douglas F. Waterhouse and was published in the Daily Bulletins of the Congress on August 20, 21, 22, and 24 1984.

The award has been sponsored by the *International Journal of Insect Morphology & Embryology* (IJIME) and its publisher the Pergamon Press, Oxford, England and will consist of U.S. $1,500 in cash and a gold medal.

The Selection Committee shall consist of 3 members of the Editorial Board of the IJIME, one representative of the Council, and the Chairperson (organizer) of the Morphology and Embryology section (or other appropriate section) of the immediate past Congress. The Editor-in-Chief of the IJIME shall be the Chairperson of the Selection Committee.

Two awards will be presented at the next Congress in Vancouver, Canada: one retroactively for 1984 and the other for 1988. Full details of the selection procedures will be published in the December 1984 issue of the *International Journal of Insect Morphology & Embryology* and subsequently in other journals.

For additional information regarding this award, one may call (201–932–9873/9459) or write to A. P. Gupta, Editor-in-Chief, International Journal of Insect Morphology & Embryology, Department of Entomology & Economic Zoology, Rutgers University, New Brunswick NJ 08903, U.S.A.