THE CONTROL OF MOSQUITOES BY PATHOGENIC MICROORGANISMS

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The control of mosquitoes by pathogenic microorganisms is just one segment of the biological control method of combating insect pests with parasitic microbes. Although the concept of microbial controls is not new, and living pathogens of insects have been tested in the field against a variety of agricultural pests in various parts of the world for many years with very promising results, the application of microbial control to vector species is only now being recognized. Most of the basic problems involved in the use of microorganisms for mosquito control work are common to the subject of microbial control in general; however, each kind of pathogen and host presents its own problems as well.

The basis for accepting microbial control as a method for mosquito abatement worthy of serious consideration lies in the knowledge that mosquitoes are attacked by many naturally occurring pathogens. Workers have frequently observed populations of mosquitoes which were decimated by diseases. Muspratt (1946) reported having sampled a larval population of *Anopheles gambiae* Giles in Africa with about 95 percent of the individuals infected with a fatal disease caused by a fungus (*Coelomomyces*). Kudo (1924) noted that protozoan parasites belonging to the order Microsporidia are commonly found in mosquitoes. He frequently found larval populations with infection rates of about 5 percent, while a population of *Anopheles crucians* Wiedemann was observed in Georgia with over 50 percent of the larvae infected. Similarly, Weiser (1956) observed populations of *Aedes* and *Anopheles* in Czechoslovakia with about 5 to 10 percent of the larvae infected with microsporidians.

Observations by the writer in California during the summer of 1959 indicate that microsporidian parasites of at least some species of mosquitoes are evidently extremely common. Larval populations of *Culex tarsalis* Coquillett, for example, were sampled over a six month period in a wide variety of habitats in the San Joaquin Valley and adjacent foot-hills of the Sierra Nevada Mountains, and infection rates ranging from about 5 to 20 percent were almost invariably found. Other species found infected included *Culex peus* Speiser, *C. apicalis* Adams, *Culiseta incidens* (Thomson), (Fig. 1), and *Anopheles franciscanus* McCracken. Infected larvae of these species usually succumb to the infections before pupation occurs, while less than 3 percent of the individuals observed in the laboratory survived to the adult stage. Presumably similar rates of infection occur in many species of mosquitoes throughout the United States. It is perhaps significant that despite the great interest in mosquitoes in California, there was only one prior record of diseased mos-

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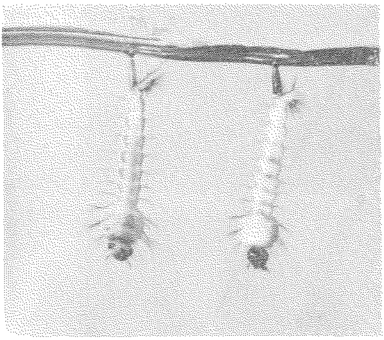

FIG. 1.—Fourth instar larvae of *Culiseta incidens* (Thomson). The larva on the right is infected with a microsporidian parasite belonging to the genus *Thelohania*.

quitoes in the state (Steinhaus, 1951). This reflects our lack of information about mosquito diseases, which is due, in part, to the difficulty of recognizing infected larvae in the field. No doubt many mosquito populations succumb to natural diseases; to the casual observer, however, their disappearances are credited to unknown causes. Unquestionably, the majority of mosquito pathogens are still unknown, while practically nothing is understood about the factors which favor the establishment of diseases in the field or the means by which their abundance can be promoted.

In the past, workers in the field of microbial control have directed most of their attention toward the study and utilization of bacterial and fungal parasites; this, however, is not an indication that these two groups of pathogens are the most abundantly represented agents of insect diseases, but merely a reflection that the two groups have had the benefit of more thorough investigation by specialists. Bacteria and fungi have known characteristics which make them easily manipulated under laboratory conditions, and have the advantage of being relatively easy to cultivate on artificial media. Moreover, they are generally considered ideally suited for purposes of microbial control, as they usually develop resistant spores, which can be produced in large quantities in the laboratory and stored for future use in the field. The development of resistant spores is highly desirable, as they can be formulated into dusts and sprays which can be applied in the field with conventional equipment.

Protozoan parasites also have resistant stages, while viruses can be stored in the dry state like bacteria; however, these two types of pathogens, because of our incomplete knowledge of their growth requirements, have not yet been cultivated artificially. In order to develop large quantities of these organisms for field tests it has been necessary to rear them in living colonies of their natural hosts. Whether it will be possible to rear these agents away from living tissues in the near future is problematical; rearing them in living hosts, however, presents no serious difficulty if their use in microbial control demands it.

More important is the problem of the infectivity of the resistant stages of the various mosquito pathogens once they have been exposed to hosts in the field. Pathogens occurring naturally in a habitat usually infect only a relatively small percentage of the host population, even though it is evident that certain disease agents are abundantly available. Epizootics, however, have been observed to develop very suddenly, leading to the devastation of entire populations; this presumably indicates that some mosquito populations are uniformly susceptible to disease, but conditions which render the pathogens infective or make the hosts susceptible occur infrequently.

As far as is known, all mosquito pathogens applicable to microbial control are one-host parasites, and infections are typically initiated through the alimentary canal. Presumably all new infections originate from spores which have been deposited in the habitat from the bodies of hosts which have succumbed to the disease. Undoubtedly only a very small percentage of the spores available in a habitat are ingested by suitable host species, and probably only a fraction of these spores is infective.

The precise conditions favoring the transmission of diseases to new hosts are apparently available to only a small percentage of the pathogens produced in nature. The release of the pathogenic agent from the resting spore stage is a critical step in the history of an infection; the physicochemical conditions of the host's gut are no doubt very important and stimulate the pathogen to terminate the quiescent stage. The limited information at hand, however, indicates that spores, particularly those of fungi and protozoa, frequently require a stimulus or maturation period external to the host; this must take place before the spores can be activated to attack the host's tissues after being ingested.

Workers have investigated the infectivity of mosquito pathogens by exposing healthy colonies of larvae in the laboratory to spores of fungal and protozoan pathogens; results of such tests have almost always been negative. These failures emphasize the difficulties of working with pathogenic microorganisms and our lack of knowledge of how the agents are transmitted in the field. The writer has exposed healthy mosquito larvae to soil and water from areas in which larvae naturally infected with microsporidians had been collected. Infections in over 10 percent of the exposed larvae have been obtained in this way. Exposure of larvae to fresh spores, however, has given only negative results. Evidently, therefore, the spores obtained with soil from the field which initiated laboratory infections had been exposed to conditions which rendered them infective. Further tests were conducted by exposing larvae to such spores which had been dried and rehydrated; these have given variable results and indicate that perhaps the initial drying of the spore is at least one step in the process. It is evident that an understanding of the process would perhaps place us in a position to encourage epizootics.

The important question is, of course, whether or not it is reasonable to expect that microbial control will ever be developed as an economic method for combating mosquitoes, or will the pathogens prove only of academic interest? It is evident that mosquitoes are attacked by the same general kinds of pathogens as have been used successfully against agricultural pests, i.e., bacteria, fungi, and protozoa. Viruses which are pathogenic to mosquitoes have not been convincingly demonstrated, but it seems likely that they also occur. Since microbial control has been accepted as an economic control method in agriculture, there appears little doubt that it also has a future in the control of pests of public health importance.

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