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California Mosquito and Vector Control Association, Inc.

Volume 50 **April 18-22, 1982**

PRESIDENTAL ADDRESS[1]

Richard C. Axtell

1982 A.M.C.A. President
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To accept the duties as President of A.M.C.A. is a humbling and challenging step. It is difficult to step into the position held throughout the past years by so many distinguished, outstanding mosquito workers. I intend to do my very best to advance the association a step closer to its long-term goals.

A.M.C.A. is a unique organization. One reason is that it is an association with a mixture of operational, policy, research - both applied and basic, and commercial people. All join together around a common goal - the best vector control possible for the enhancement of the quality of human life while wisely managing the environmental resources. All of these people contribute their particular talents enthusiastically so that collectively the welfare of people is improved. A.M.C.A. is a splendid example of people joining together and putting aside minor differences to accomplish a common goal.

I hope my diverse background will enable me to be sensitive to all facets of our diversity in A.M.C.A. I am a New Yorker who has lived for 20 years in North Carolina, so I have experienced and appreciate differing viewpoints. I have been in mosquito control field development research for many years in North Carolina and in much earlier times (in the 50's) in the army in Maryland. I have spent time in screening and testing insecticides as an employee of a chemical company. In my younger more carefree days, I taught and did applied field work in insect control in the Philippines. In the years at North Carolina State University, I have taught medical and veterinary entomology and conducted research, both applied and basic, on related pest problems - namely, mosquitoes, tabanids, *Culicoides*, ticks, eye gnats and filth flies. I must admit, however, that I have never done taxonomic work on mosquitoes - never revised a group or described a new species. For this I am sure our true taxonomic members are grateful! I think that this shows that I do have enough common sense to recognize when I am not competant.

Another reason that A.M.C.A. is unique is that it is both national and international. A.M.C.A. works with and through state and regional associations in many ways - most conspicuous is our Annual Meeting held traditionally with a local host association. We help each other and together provide a national strength to promote professional mosquito control. More and more we have members outside North America and are looked to for leadership in mosquito and vector control. I plan to promote in my term the strenghtening of these international bonds. This will be of mutual benefit. I had the opportunity to participate in the regional WHO Seminar on Integrated Vector Control in Adana, Turkey, last November and we found a great desire and need for closer international ties. We all share common goals and technology regardless of regional or national loyalties. We are living in changing times. Human population growth and economic development are basic world problems. Mosquito control is increasingly sophisticated. It is understating the case to say that we have had the technology for a long time. Some of it - yes, if we are just talking about killing mosquitoes. But more and more we are dealing with the triad of Mosquitoes-Humans-Environment. The members of A.M.C.A. are concerned with all three. Human behavior and economic development often rapidly change the environment and increase the threat from mosquitoes. People spoil their nest! You know the worldwide examples, such as irrigated cropland, massive reservoir

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[1] Presented at the 50th Annual Conference of the California Mosquito and Vector Control Association at the Capital Plaza Holiday Inn, Sacramento, California, April 18-22, 1982.

projects, waste disposal lagoons, solid waste disposal. I happen to work on the problem of organic pollution and tremendous production of *Culex quinquefasciatus*, such as from animal waste lagoons, and on *Aedes* production from manmade coastal dredged material disposal areas. We have several people in A.M.C.A. concerned with *Anopheles*, *Culex* and *Psorophora* population explosions from irrigated riceland and pastures. Human behavior and economic development are creating problems faster than our technology and our financial resources can solve them. Yet at the same time, there is a decline in interest and funding of operational and research work on mosquito control. The true cost of economic development is being underestimated because the ultimate costs in human disease and needed mosquito control are not being recognized.

A.M.C.A. and its members are facing a real challenge in conveying to the citizens and the policy makers that we do not just deal with killing mosquitoes. Rather we deal with the triad: Mosquitoes - Humans - Environment. As a corollary we have to increase our professionalism, training and sophistication of our technology. We have many people in A. M.C.A. who have mastered the art and science of mosquito control very well! Others need help. We are moving to establish regional A.M.C.A. - C.D.C. joint training courses to help meet this need for better training.

All in all, A.M.C.A. has an illustrious past and a bright future. If you will all promote the A.M.C.A. by bragging about it at every opportunity and bringing in new members, both national and international, we will be even stronger and listened to more. Together we will flourish and better serve mankind. I am gratful for the opportunity to serve you and the A.M.C.A. in the coming year.

A MEMORIAL TO FINLAY, REED, GORGAS AND SOPER AS MAJOR CONTRIBUTORS TO PRESENT DAY CONCEPTS ESSENTIAL FOR CONTROL OF MOSQUITO-BORNE ARBOVIRUSES

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It is a privilege to memoralize the contributions that Carlos Finlay, Walter Reed and William Gorgas made to our understanding of the epidemiology and control of diseases caused by mosquito-borne viruses. As you will see, I have added Fred Soper to this distinguished group. I also took this opportunity to review our present knowledge of vector biology that is critical to the control of epidemics of these diseases in North America.

First, let me review the significance of the early contributions of the above pioneers in this area of science.

The first major discovery relevant to the control of arboviral diseases was in 1900 when Reed (1901, 1902) reported on the Yellow Fever Commission's studies in Cuba. The studies were based in large part on the observations and hypotheses developed by a Cuban, Carlos Finlay. The Yellow Fever Commission reported that they had transmitted yellow fever from man to man by the bite of a mosquito *Aedes aegypti*. Gorgas (1911) quickly applied these findings in a program for control of *Ae. aegypti* and successfully eradicated yellow fever from Cuba where it had prevailed for over 150 years. Control of this major mosquito-borne disease depended on the finding and removal or treatment of almost every breeding source of the vector in the domestic environment. The principal was to reduce the vector population below the threshold level essential to maintain the viral transmission cycle.

Subsequently, eradication of *Ae. aegypti* became the focus of a major program of the Rockefeller Foundation led by Fred Soper (1943). For practical purposes yellow fever was controlled and eradicated from major urban centers of the World where it had prevailed for over a century. However, the discovery of jungle and rural cycles of yellow fever in the Americas and Africa (Strode, 1951) put a damper on the hopes of yellow fever eradication and explained the reappearance of virus in urban centers where it had been eradicated. Subsequently, an effective vaccine was developed (Thieler and Downs 1973) and this provided an alternative approach to protect rural populations in areas where vector control was impractical.

The point of the preceding brief historical review was to emphasize that for over 80 years we have understood the epidemiological factors that control the spread of a major mosquito-borne viral disease. Studies over the past 40 years have extended this knowledge to a wide array of other arboviruses including the demonstration that prevention and control of epidemics caused by such agents is feasible by vector control although it is expensive.

My concern is that permanent and effective control programs for mosquito-borne viruses are non-existent in most areas even though we continue in a most impressive fashion to add to our epidemiological knowledge of the diseases associated with mosquito-borne viruses and have extended our degree of sophistication regarding the vectors and the causative viruses.

The above background on yellow fever in its urban cycle serves well for an extension of the discussion to dengue fever. It has been known since 1902 (Graham 1903), that dengue fever was a viral infection transmitted in an urban cycle by *Ae. aegypti*. In recent years it was found that four distinct viruses can cause the disease; that the infection spectrum goes from inapparent infection through febrile illnesses to hemorrhagic fever, shock syndrome and death; and that there are alternative vectors to *Ae. aegypti* particularly in the Pacific area. None of these important findings have changed the basic concept that developed from the yellow fever studies, namely, that reduction of the vector population to levels that cannot support the transmission cycle is the most feasible approach to control. In spite of this knowledge, the dengue fevers continue to occur at endemic or epidemic levels or are reintroduced repetitively over extensive areas of Asia, the Pacific and Caribbean Islands, and South and Central America. As a further expansion of this concern, an *Ae. aegypti* control and eradication program was making excellent progress in the United States in the 1960's (Smith 1967) when our government decided that it was costing too much and wasn't that important. The annual budget in 1968 was 16 million dollars and 2,610 persons were employed. For practical purposes, the program was abandoned within two years. Major urban centers in the Southeastern United States now report that *Ae. aegypti* is a major pest and I assume that these are potentially receptive areas for dengue and yellow fever.

Until recently the occurrence of dengue was referred to as a problem of the underdeveloped areas of the World. If this is the case, we can now include parts of Texas and Queensland, Australia in that classification as dengue virus was reintroduced into those areas and they were receptive as there was an adequate population of *Ae. aegypti* to support transmission and there was a susceptible human population.

Arbovirus epidemiologists were not surprised and had anticipated these developments. The point is that there was no concerted effort to carry out the principles of vector control that were established by Gorgas and Soper. A dengue fever epidemic was reported recently in Cuba with over 300,000 cases and 158 deaths. This could not have happened in the early 1900's at the time of Gorgas' successful vector control program. The response of the Cuban government is noteworthy and is reported in detail in the Epidemiological Bulletin of the PAHO (Anonymous 1982). The Cuban Government has established a program that can serve as a model for complete response which includes island-wide house-to-house search for and eradication of *Ae. aegypti* which is being supplemented by island-wide applications of organophosphorous insecticides. I recommend the above report as essential reading. Few countries in the Western Hemisphere are prepared to meet the inevitable high cost of such a program today. I would add that dengue virus infections have remained endemic in Puerto Rico and Southeast Asia since the 1960's in spite of any efforts to establish control programs.

I have expressed concern previously (Reeves 1972, 1980) as have others (Downs 1981), that areas where dengue viruses prevail are potentially receptive to yellow fever. A recent conference on yellow fever (Woodall 1981) reiterated this concern and recommended the use of 17D vaccine in the event of an epidemic. However, further study revealed that only seven million doses of the vaccine were available in the World and that this supply probably could not be doubled in the event of an epidemic. That amount of vaccine will not control yellow fever if it is introduced into major urban centers in the Americas, Asia or Africa. Similarly, there is little chance that effective vaccines for other arboviruses will be available in large amounts in the near future.

I do not believe that the failure to control dengue or yellow fever is for lack of knowledge on the populations at risk, the causative viruses or which mosquitoes are vectors. In February of this year I presented a paper (Reeves 1982) at the International Seminar on Viral Diseases in South-East Asia and the Western Pacific on the expanding gap between the epidemiological knowledge of arboviruses and their effective control. I concluded that vector control programs were ineffective because of:

1. A primary dependence on insecticides rather than source reduction for vector control.
2. Legal restrictions on the use of insecticides and lack of a legal basis for water resource management in both urban and rural environments.
3. A limited knowledge of vector behavior that is essential for control of the adult female population that is transmitting infection.
4. A low priority by political bodies for funding of control programs due to lack of belief by the public that there is a need for action until an epidemic is in progress or a pest population is out of control.
5. An acute shortage of adequately trained persons to investigate the problems and initiate effective control programs.

I believe that these comments are equally applicable to control of the mosquito-borne viral diseases that prevail in North America and that concern the majority of this audience. What are my thoughts on the problem?

Western equine encephalomyelitis (WEE), Eastern equine encephalomyelitis (EEE), St. Louis encephalitis (SLE) and representatives of the California encephalitis (CE) complex are the principal mosquito-borne viral diseases of concern in North America. On an international scope we could add Japanese encephalitis, Murray Valley encephalitis, West Nile, Venezuelan equine encephalitis and other diseases as examples. These infections differ from yellow fever and the dengues in that they rarely or never are spread from man to man by a vector but rather depend on transmission between wildlife hosts or transovarial infection in their vectors for their basic maintenance. Spread to man is an accidental event and of no importance in viral perpetuation. Regardless of that, the principles developed by Gorgas and Soper apply– reduce the vector population to threshold levels below that required for effective transmission if you wish to prevent epidemics. To accomplish that objective, requires the establishment of a long-range program of vector abatement backed up with a surveillance system and a capacity to act rapidly when environmental conditions provide a warning of impending epidemics that will require emergency abatement of large adult vector populations.

Detailed epidemiological studies of WEE and SLE in California (Reeves and Hammon 1962) provided a basis for development of a state-wide surveillance program in California in the 1960's. Similar programs each tailored to fit the epidemiological variations of the different mosquito-borne viruses that prevail, are now established in at least 18 states and several provinces in Canada. The Vector-Borne Viral Diseases Division of the Centers for Disease Control summarizes information that is gathered into their Encephalitis Surveillance Reports. The six types of information that represent the core content of an arboviral surveillance program (Reeves and Milby 1980) are knowledge on:

1. Water availability from precipitation and other water resources that are available for vector production.
2. Occurrence of temperatures that favor or disfavor development of vector populations and viral development in those populations.
3. Monitoring the levels of the primary mosquito vector populations.
4. Viral activity in:
   a. Vectors
   b. Sentinel hosts
   c. Clinical cases
5. The economic manpower and equipment resources available for routine and emergency vector control programs.
6. Assessment of the probable effectiveness of alternative programs to control the adult female vectors that are infected and are transmitting infection.

It must be emphasized that surveillance is based on information gathered at and used by agencies concerned with control at the local level. Centralized reporting and services at the state or national level only serve to spread knowledge and to extend the system to a broader geographical area.

The experiences in Kern County, California in 1958 (Reeves et al. 1964) Dallas, Texas in 1966 (Hopkins et. al. 1975) and Manitoba, Canada in 1981 demonstrated how impending or in-progess epidemics of WEE and SLE can be attacked and how important a surveillance program can be. There is no reason to question that the emergency control of vector populations in these and other epidemics had a desirable effect. At the same time I believe that we learned certain lessons from these experiences.

When emergency control programs have to be instituted in an epidemic, the cost of human suffering is already high. There were 15 WEE and 2 SLE cases in Kern County, 145 cases of SLE in the Dallas epidemic and 25 cases of WEE in Manitoba. Cases will have occurred and other individuals will be in the incubation period when the emergency program is started. Such programs are expensive-- almost $2,000,000 was spent to apply ULV Baygon by air over an area of 450,000 hectares (over 1,000,000 acres) in Manitoba. Programs can also be ineffective because of insecticide resistance of the vector or delays in action by endless debate over the health implications of insecticide exposure or regulations that limit the use of effective insecticides. However, we have come a long ways from the 1950's and 1960's as we now know that control of the adult female vectors that are infected and are transmitting infection is the only measure that will immediately abate an epidemic of WEE or SLE. ULV aerial application of insecticides is the current method of choice to control such epidemics. Elimination of the infected and infective mosquito population over a large geographic area for the 4-5 day period of viremia in vertebrate hosts will eliminate the etiological reservoir and prevent new infections in the vectors and transmission to humans.

I want now to identify some areas of research on the biology of vectors of arboviruses that will further our knowledge of the epidemiology of these infections and improve programs to control the diseases. In the interest of time, I will present concepts and problems rather than detailed data.

The ovaries of female mosquitoes contain markers that allow us to separate nulliparous from parous individuals. These markers can be used in combination with fluorescent dusts in mark-release-recapture studies to make estimates of adult survivorship and population levels (Milby 1979, Nelson et al. 1978, Nelson and Milby 1980). The resulting life tables have contributed to our understanding of the dynamics of viral transmission and led us to fully appreciate that the older viral transmitting females are relatively few in the population and should be the primary target for control in the event of an epidemic.

However, there is a gap in our knowledge if we wish to implement such programs. I do not know of any current in-depth field research to determine if the usual ultra-low volume applications of insecticides for adult mosquito control are equally affective against parous and nulliparous females, old versus freshly emerged females, or females derived from insecticide resistant versus susceptible larval populations. Such data are essential if the objective of epidemic control is to eliminate the older adult vector population and interrupt viral transmission.

Tests of mosquito pools for viral isolations is a most important aspect of surveillance programs. However, little attention has been given to the collecting techniques for such pools. A method may be selected because it is easy and will provide large samples rather than be a technique that will assure that the samples contain significant numbers of the feeding parous females that can be infected and transmit virus. There is little purpose in testing large numbers of freshly emerged nulliparous females unless one is looking for transovarially transmitted viruses.

If a vector control program is to be effective and epidemics are to be controlled, it is essential that the primary vector species be identified for each virus in the area of concern. We used to assume that the isolation of virus from a species was sufficient reason to condemn a species as a vector. We now know this is not so as studies on vector competence (Hardy et al. 1979) have shown that many species that feed on infected vertebrate hosts will ingest virus but are ineffective vectors. A significant number of even the most efficient primary vector species can be infected with but never transmit viruses effectively. The following experiences illustrate the problem. We have observed that the levels of WEE viral transmission can remain low or be undetectable in a *Cx. tarsalis* population that has risen to a high level over an extensive area (Reeves 1970). This was contrary to our earlier epidemiological experience and required an explanation. We also had observed that in years of high levels of viral transmission an average of only one in four *Cx. tarsalis* infected with WEE virus could transmit infection (Reeves et al. 1961). It seemed unlikely that a failure to have completed the extrinsic incubation period was a sufficient explanation for these observations.

Studies have revealed there are wide variations in the vector competence of *Cx. tarsalis* subpopulations for WEE virus (Hardy et al. 1976). Subpopulations have been selected from a single colony that are either highly resistant to or highly susceptible to WEE viral infection and resistance is a recessive genetic trait (Hardy et al. 1978). To our surprise, many *Cx. tarsalis* that became infected when fed on low titer viremias never transmitted infection by bite even though the virus multiplied to high levels after incubation for over 21 days at high temperatures. These studies revealed that there were dose-dependent gut and salivary gland barriers that limited vector competence (Kramer et al. 1981). The studies are now extended into very sophisticated evaluations of the influence on viral infection of cell receptors, cell membranes, enzymes, inhibiting substances in haemolymph and various temperatures of incubation in the vector.

Extrinsic incubation temperatures have profound effects on the growth of different viruses in their vectors. It is interesting that no studies on extrinsic incubation of arboviruses have been done in climate chambers that duplicate the fluctuating temperatures where vectors live. Such studies could lead to exciting and unexpected results.

Dr. Hardy and associates are studying the interesting question of why *Cx. tarsalis* is the primary vector of SLE virus in the Western United States while the *Culex pipiens* complex, although abundant in the same area, is a secondary vector at best. In contrast, in other parts of the United States the *Cx. pipiens* complex is a primary vector of SLE virus during epidemics (Monath 1980). Vector competence studies indicate that *Cx. tarsalis* is an efficient vector of SLE viral strains collected from a wide range of areas in the United States. It has few salivary gland barriers to efficient transmission. In contrast, *Cx. pipiens* from California is an inefficient vector of California viral strains as compared with *Cx. tarsalis*. There are several hypotheses to explain these differences which are under study. The significance is that with confirmation we will be able to concentrate vector control programs in California and other western areas on the competent *Cx. tarsalis* even though *Cx. pipiens* are abundant.

Field studies on the life table of adult female *Cx. tarsalis* combined with the studies on vector competence have made us realize how tenuous viral transmission cycles can be. We find for *Cx. tarsalis* that the fertility rate of females, survival of immatures and resulting numbers of adult females is quite low as compared to the average of over 150 eggs deposited per female at each oviposition. Once an adult female emerges she does not mate until 24 to 48 hours of age. If autogenous, she delays the first blood-meal until oviposition of those eggs four to five days later. Her first possible contact with a viremic blood-meal is at three to six days of age and the majority of such meals will not be viremic. Meanwhile, daily mortality or losses from the resident vector population by dispersal may range from 25 to 40% per day (Nelson and Milby 1980, Nelson et al. 1978). Even if infected with virus, a surviving female probably will oviposit and seek the next blood-meal within five to seven days at which time she may not have completed extrinsic incubation so she cannot transmit infection. She must take a third blood-meal and be a competent individual who has passed virus in infectious quantities into the salivary glands and the host must be susceptible to infection or the cycle is broken. This biological situation is so tenuous that you must wonder how arboviruses persist and why they are so hard to control, but they do. However, this background increases our confidence that detailed knowledge of the interaction of biological variables is the basis for planning the most efficient vector control program. The older viral transmitting female vector populations are clearly identified as the primary target in the event of an epidemic. An effective adulticiding program should increase daily mortality to over 90% which should stop the viral transmission cycle.

I could extend this discussion to consider the importance of genetic studies on vectors, other types of biological studies and statistical modelling of vector populations but time will not permit. Sufficient to conclude this section with my belief that many such areas can still be studied profitably to improve control programs and epidemiological studies. We have a wide array of new techniques that can contribute to the studies. Until recently, it has been relatively easy to obtain funds for basic laboratory research on vectors and to train students and staff, but this is no longer true. It has always been and will continue to be difficult to obtain adequate funding to put into operation and maintain effective control programs. So, I reiterate the point I made at the opening of this paper, that we have the basic tools to extend our epidemiological knowledge and to implement effective control programs but must continue to develop more complete knowledge and increased control efficiency in those programs.

I believe that we must all make an effort to influence the public and its elected officials to utilize present knowledge for effective control of arboviral diseases. Such action is a most fitting memorial to the contributions of Finlay, Reed, Gorgas, Soper and their successors.

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INTERNATIONAL DEVELOPMENTS IN THE CONTROL OF INSECT VECTORS OF DISEASE

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ABSTRACT

Chemical pesticides have been the foundation of agricultural and public health pest control for several decades and are likely to remain the mainstay for the control of vector-borne diseases in the foreseeable future. To combat the accelerating development of pesticide resistance in insect vectors of human diseases, which has been exacerbated by the heavy use of agricultural chemicals, the World Health Organization (WHO) has been carrying out a comprehensive program of research on vector biology and control in collaboration with its Regional Offices, the Member States and through Collaborating Centers. The program is aimed at understanding the ecology and population dynamics of vectors, evaluation and testing of new insecticides and new techniques of application, preparation of countermeasures to the insecticide resistance problem, the search for and development of biological control agents for control and integrated control of the important insect vectors of disease. WHO has also stressed that the active participation of the people and the community is essential if abatement and control of disease vectors are to be effective and economically achieved. This paper presents a brief resume of recent developments which should help to dispel some of the pessimistic views that have been expressed. Certain trends that have emerged during the past few years may be pointers to the future and are briefly discussed.

INTRODUCTION.–During the past 35 years some spectacular successes have been achieved in the control of certain vector-borne diseases such as typhus, plague and malaria, but the recent resurgence of malaria and the increase in some of the vector-borne diseases have presented some problems. On the other hand, very successful ongoing programs, e.g., for the control of onchocerciasis in West Africa, indicate that other vector-borne diseases such as filariasis, trypanosomiasis, Chagas' disease, leishmaniasis and schistosomiasis could also be brought under more effective control. However, sometimes concern has been expressed, for example, "We are at crossroads today in our ability to control these diseases. We have relied on chemical insecticides as a major method of vector control, but growing problems of vector resistance to the insecticides, their rising costs and major concerns regarding their environmental impact have introduced an element of

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uncertainty and technological despair."[1] There is no doubt that alternative strategies are required for long term solutions to the problem because the need for vector control, particularly in developing countries, remains undiminished. Prevalence of some of the vector-borne diseases in the world is given in Table 1.

Increasing emphasis is therefore now being placed on research and field trails of alternative methods of vector control. The World Bank/UNDP/WHO Special Program for Research and Training in Tropical Disease has established a scientific working group to support priority research on biological control. This group will further augment the efforts of WHO's Vector Biology and Control Division which has already been looking into problems of insecticide resistance as well as alternative methods for the integrated control of insect vectors of disease.

Concurrently with these technical developments, far-reaching changes have taken place in the health polices of the world. In 1977 the World Health Assembly decided that the main social target of governments and WHO would be the attainment, by all the people of the world by the year 2000, of a level of health that will permit them to lead a socially and economically productive life, popularly known as "Health for All by the Year 2000". In 1978, an international conference on Primary Health Care[3], held in Alma-Ata, USSR, stated that primary health care was the key to attaining this target. In 1979, the World Health Assembly launched the Global Strategy for Health for All when it endorsed the Alma-Ata Report and Declaration and invited Member States to act individually in formulating national strategies and collectively in formulating regional and global strategies. This global strategy was adopted by the World Health Assembly in 1981.[4] The strategy is based on the concept of countrywide health systems based on primary health care. The strategy involves specifying the measures to be taken by individuals and families in their homes, by governments, and by the health services at the primary and supporting levels. It also involves selecting technology that is appropriate for the country concerned in that it is scientifically sound, adaptable to various local circumstances, acceptable to those for whom it is used and to those who use it, and maintainable with the resources the country can afford. Crucial to strategy is making sure of the social contents of the health infrastructure and technology through a high degree of community involvement.

Thus, for example, the global strategy for malaria control envisages the delegation of responsibility and resources to the communities so that where possible they will carry out agreed components of the health program such as insecticide spraying. Operational problems arising from the socio-economic context within which the control of vector-borne diseases has to be carried out include the following: (a) limited financial resources, especially the availability of hard currencies to the developing countries: (b) very often limited trained manpower, especially in the peripheral rural areas; (c) sometimes low population densities over a wide area; (d) in some areas, difficulties in access during part of the year because of heavy rainfall; and (e) the repetitive nature of vector control measures, etc.

CHEMICAL CONTROL.–WHO has operated a program for the evaluation and testing of new insecticides for about 20 years. Up to now, about 2000 compounds have been evaluated for their effectiveness against disease vectors and safety to man. Unfortunately the number of new compounds entering the scheme has decreased progressively in recent years.

For example, in 1978 no new compounds were received and in 1979 only six compounds were submitted for testing. The industry has, however, recently been approached anew and it is hoped that new compounds and new groups will be forthcoming. The method usually followed to countereact insecticide resistance is to change to another insecticide. How-

Table 1. Prevalence of some of the vector-borne diseases in the world.

Disease	Carriers	At risk
malaria	400,000,000	one to one and a half billion, maybe more
lymphatic filariasis	50,000,000	250-500,000,000
schistosomiasis	200,000,000	500,000,000
onchocerciasis	20,000,000	100,000,000
African trypanosomiasis	a few thousand	100,000,000
Chagas' disease	12,000,000	about 24,000,000
yellow fever	a few hundred	⩾100,000,000
plague	a few hundred	500,000,000

ever, this approach is only possible if alternative insecticides are available. It seems clear that a better lesson should be learnt from our current knowledge about the development of resistance, i.e., a deliberate policy on usage should be introduced to ensure that the usefulness of an insecticide will be maintained over the longest period possible. Among the parameters that have been examined in simulation experiments are the relative effect of the dose of the insecticide employed, the application and selection thresholds, altering selection/relaxation schedules, residual vectors which permit the preservation of genes that confer susceptibility, lower reproductive fitness of resistant genotypes, the influx of susceptible migrants, and the varying decay rates of insecticide resistance. Certain proposals for possible control methodology[5] have emerged from these theoretical studies on the dynamics of resistance such as:

(a) Dosage management and the control of gene dominance: with residual insecticides it is clear that selection and evolution of resistance is affected strongly by the toxicity of the residues towards the heterozygote. However, since chemical residues are subject to weathering, the selection is continuously modified by the rate at which the insecticide residue tends to disappear. Resistance evolves faster with more persistent chemicals and is delayed with the less persistent chemicals, depending upon the migration of susceptible individuals. It would seem that the prolonged stability of residues, while advantageous economically, might in fact be detrimental to the speed of development of resistance.

(b) Mosaic application of chemicals: reduction of insecticidal selection pressure by continual dilution of the surviving segment of the population as a result of mutual population exchange due to natural dispersal. The mosaic envisaged would normally employ two or three unrelated insecticides, thus maintaining overall insecticide coverage of the operational area.

(c) Mixtures, rotation and optimum sequential usage of chemicals: some of the theoretical papers published on the subject suggest that where chemicals with contrasting modes of action and detoxication pathways are employed, a delay in the onset of resistance might result. Insects that may survive one of the chemicals are killed by the other. At a recent meeting of the European Society of Nuclear Methods in Agriculture (1981), the evolution of resistance/control strategies was discussed. The question of whether isolation is to be preferred to immigration was thought to be a complex matter which required further clarification. There was also discussion about what constituted fitness of R gene. However, assuming equal reproductive fitness, what is important is whether an insect lives or dies after the insecticide is applied. Low dosage is preferred because this exerts a low selection-pressure. Survival of heterozygotes is particularly important in an already resistant population as otherwise the susceptible gene will be lost in the population. Mani and Ward stressed the importance of killing heterozygotes by ensuring a high insecticide pick-up.[6] They also pointed out the need to ensure that a proportion of all genotypes will be able to escape contact with the insecticide altogether.

Although the above aspects of control methodology have been studied and discussed, field experiments have only recently been started and the results are awaited with considerable interest to see whether these will provide a practical solution to the resistance problem. Formulation of satisfactory operational approaches to the application of insecticides remain[5] one of the most important problems of vector control.

BIOLOGICAL CONTROL.—For more than 20 years WHO has considered the use of pathogens, parasites and predators of vectors but, owing to lack of funds a major research program in this field was developed only 10 years ago. This program involves (a) the search for new promising agents; (b) the assessment of their potential for controlling key vector species and their safety to mammals and in the environment; (c) studies on the possibility of their mass production; (d) the stability of the formulated product during storage and under different methods of application; and (e) eventually the epidemiological evaluation of the effect of these biological agents during disease control operations. A brief description of progress made so far is given below in respect of a few of the agents.[7,8,9]

Bacteria. For many years *Bacillus thuringiensis* has been used against certain crop and forest pests. Until 1977, all the known strains of this bacillus were specific only for caterpillars. Thus, it created great interest when in 1977 Dr. H. de Barjac identified a new serotype, H-14, which was highly pathogenic to mosquito larvae. This strain had been found in Israel a month earlier by Dr. Goldberg and Dr. Margalit. It is now known to be effective against the larvae of most mosquito species as well as blackfly larvae. It also has the great advantage of being specific to these dipteran families, which means that this serotype does not affect non-target organisms and is not harmful to man. The stability of industrially developed formulations of serotype H-14 has been found to be satisfactory when they are stored under tropical conditions. H-14 is, therefore, highly suitable for the production of a non-residual bacterial larvicide with a great safety margin for man and the environment, and is already being mass produced for operational use.

Strain 1593 of *Bacillus sphaericus* has greater effectiveness against the *Culex* species that breed in polluted waters. It has a narrower spectrum of effectiveness than *B.thuringiensis* H-14 but could be used in the production of a bacterial larvicide for *Culex* and possibly anopheline control. It is also a good candidate for local production in small or cottage industries. The effectiveness of such bacterial agents can perhaps be further improved so that an adequate concentration of the material produced by the bacteria will remain for longer periods in the same water layer as the mosquito and blackfly larvae. Observations are also being made on whether these bacteria recycle, once introduced. Since bacteria can only be used as non-residual larvicides, their application will be limited; for example, it would be operationally difficult to treat extensive rice fields every 10 days or fortnightly.

Fungi. The Australian strain of *Culicinomyces clavosporus* has been found to be partially effective against mosquitoes and has been shown to be safe enough for man and the environment. *Coelomomyces iliensis* has been found in the USSR to be a satisfactory larvicide against *Culex* mosquitoes. It has a narrow spectrum of activity which may have recycling potential at operational levels under favorable environmental conditions. Fungi may also have a potential for the control of filaria and malaria vectors in permanent and semi-permanent breeding sites. However, their mass production and distribution constitute major problems at present although they may be very valuable for local usage. Because the fungus mass produces extremely resistant sporangia, the pathogen is able to exist in the environment for a long time, causing other epizootics when susceptible intermediate and/or definitive hosts appear. Natural spread of the pathogen without industrial or laboratory production into new adjacent areas and its artifical distribution by scientific workers into other regions including rice fields have been noted. Other species which attack anopheline populations have been reported from the Philippines and India.

Protozoa. Two microsporidian species, namely *Nosema algerae* and *Varraia culicis* exhibit a high pathogenicity against mosquito larvae and can interfere with the ability of mosquitoes to be infected by the malaria parasite. However, the spores of these species do not persist for long in the feeding zone of mosquito larvae. Accordingly, further testing of these organisms will depend on the development of a formulation that will prevent the spores from sinking to the bottom of the bodies of water to which they are applied. Mass production is only possible through the proxy hosts, caterpillars.

Nematodes. It has been found that *Romanomermis culicivorax* has a wide mosquito host range, including major vectors of malaria. It is safe for man and non-target organisms. It can be mass-produced by simple means at the cottage industry level as long as cheap semi-skilled manpower is available. However, these nematodes can only be regarded as narrow-spectrum larvicides whose recycling at an operational level is uncertain. Mermithid nematodes with a better potential are therefore being investigated.

Larvivorous fish. A number of fish species have been used to control mosquito larvae, with special emphasis on malaria and filaria vectors. A review of the use of such fish since 1970 has shown that *Gambusia* has been used in 40 countries, *Poecilia* in 15, annual fish in one, and other species in 11 countries.[10] The efficiency of these fish for mosquito control has mostly been empirically determined. In some countries remarkable results have been reported, while in others the results have been somewhat disappointing. However, the use of larvivorous fish (especially indigenous species) is one of the most promising biological control methods. Additional investigations are required on epidemiological assessment and environmental safety.

Toxorhynchites species. These are predatory mosquitoes with biological control potential. Female *Toxorhynchites* search for oviposition sites such as tree holes, leaf axils, etc., where they can multiply and redistribute themselves. *Toxorhynchites* could perhaps be one method to be considered for the control of lymphatic filariasis vectors in South Pacific areas and of *Aedes aegypti* in the continent of Africa. However, mass production and storage constitute serious problems at present. They might be a good candidate for local production by the communities concerned.[11]

It is hoped that at least some of the above biological agents can soon be utilized in integrated control activities against the vector-borne diseases.

GENETIC CONTROL.[12,13,14]—During the past twenty years increasing research on genetic methods for the control of insects of public health and veterinary importance has been carried out. The basic theory of insect pest control i.e. taking advantage of their mating behavior, is well-known. This technique usually involves the release of reared insects into a natural population after they have been made either sterile or genetically altered or treated to disrupt further reproduction or make them harmless to man by selecting refractory strains. In either case, the released insects should compete with normal populations for mates, resulting in the transfer of their sterility or other genetic abnormalities to the eggs laid by wild females. Another possible approach is to attract members of the wild population to a trap where they are sterilized and then allowed to return to the wild population to compete for mates.

It may be mentioned that genetic control methods are still incompletely developed. However, control of the screwworm fly has been highly successful by using these methods over large areas of southern USA and Mexico. The field trials with insects of public health importance such as mosquitoes and tsetse flies have only been on a small scale so far and for the most part only partially successful. Therefore, appraising the future role of genetic control methods in public health is difficult at present. It is hoped that in the future methods might be developed that could be more efficiently used in an integrated program of insect control than by themselves. The major benefits of genetic control methods, if and when developed, would be selective action on the target pest and the avoidance of adverse effects on non-target organisms. Compared with insecticide spraying, genetic control methods require more biological, behavioral and ecological information about the pest.

Several different methods are being studied for the genetic control of insects. These may be classified into three broad categories:

(a) **Sterile insect release method**: this method is usually based on the use of ionizing radiation or chemosterilants to induce sterility in reared males through dominant lethal mutations. This method is so far the simplest and best developed of the proposed genetic control methods.

(b) **Release of genetically altered insects**. Release of such insects with any of the following mechanisms not only has some impact in the first generation but may continue to exert suppression in subsequent generations by infusing deleterious genetic material into a portion of the natural population. These mechanisms include chromosomal translocations, conditioned lethals, compound chromosomes, etc.

(c) Sterilization of members of the wild population: This method is known as auto-chemosterilization and would have the advantage that the expense of mass rearing of insects would be avoided. Chemosterilants seem to be the only practicable means of sterilization which could be employed at present.

Field experiments so far have been carried out on *An. albimanus. An. culicifacies, An. gambiae, Culex quinquefasciatus, Culex tarsalis, Aedes aegypti,* and tsetse flies with varying degrees of success,[15,16,17] but without complete success.

As would be evident, there are a number of operational constraints for the genetic control of the insect vectors of disease. A basic requirement for all genetic control techniques is the capability of rearing the required number of vigorous male insects in sufficient number and at a reasonable cost. Furthermore, facilities are required for their release over an extended area and a precise knowledge of the timing and frequency of release. There is little information available on the changes brought about by laboratory rearing. Much more information is needed on this point in relation to the validity of behavioral studies based on laboratory colonies. Many ecological aspects of the target species may be vital, for example, an accurate estimate of natural populations in time and space and their rate of reproduction and density dependent regulation of populations both under natural conditions and under stress. Finally, the cost-effectiveness of genetic methods of control should be determined as very little precise information is at present available on the possible cost per km^2. Genetic control methods are generally much more sophisticated and, for this reason, more skilled manpower is required. Unfortunately there is a great dearth of trained personnel available in most countries where vector-borne diseases are prevalent for implementing any form of vector-control measures.

ENVIRONMENTAL MANAGEMENT.–Environmental management had, in the past, been one of the main tools for the prevention and control of vector-borne diseases; it involved, for example, the reduction and elimination of breeding areas by means of drainage, leveling, flushing, management of vegetation and salt concentration, intermittent irrigation, etc.[18] But these measures have been relatively neglected since the introduction of residual insecticides for vector control. Only recently has it been recognized that environmental management methods should be reintroduced and applied jointly with all the available procedures for preventing vector-borne diseases (in USA their use was never stopped). These diseases are often linked to poor socio-economic conditions but some can be associated with projects that are aimed at economic development. Thus the construction of dams for power generation, irrigation, flood control, and water supply may lead to an increase in the prevalence of malaria, schistosomiasis and onchocerciasis, while human settlements may increase urban filariasis and domestic rodent-borne diseases. In 1979 the WHO Expert Committee on Environmental Management for Vector Control[19] reviewed the available measures that had been developed in the past for the control of many invertebrate and vertebrate hosts and vectors of human and animal diseases, and assessed the present application of environmental management techniques in vector control programs and the interaction of these measures with agricultural irrigation and other socioeconomic development schemes. This Committee also formulated important recommendations for improving the prevention and control of vector-borne diseases through the application of these measures.

INTEGRATED PEST MANAGEMENT.–The FAO Panel of Experts on Integrated Pest Management (IPM) in 1967 gave the following definition: "A pest management system that in the context of the associated environment and the population dynamics of the pest species utilizes all suitable techniques and methods in as compatible a manner as possible and maintains the pest population at levels below those causing economic injury."[20] Of course others have defined IPM slightly differently. However, a word about the economic injury must be said as it concerns the field of public health. In agricultural pest managemet, the assessment of economic losses or the setting of an economic threshold which is an essential part of the pest management planning process can be done with an adequate degree of accuracy. In the context of disease control, the process is much more complicated. Measurement of cost/benefits in a health project has often been questioned because it is not possible to quantify health and well-being in terms of dollars and cents. Thus, the setting up of an economic threshold for a disease is often difficult or unrealistic. The following simplified definition of integrated control in the health field has therefore been suggested: "the selection and application of methods of control to optimize the achievement of results."[21]

One lesson that has been learnt from the recent past in vector control is that no single method is available to satisfy each and every situation and that it will be necessary to combine the methods best suited to local expertise, self-help and commitments on the part of individuals and the community, and improved health education. Although some progress has been made in the pest management of agricultural[22] and public health pests, good practical examples are rather limited. Metcalf (1975) has stated that "pest management programs against the insect vectors of human disease are perhaps more intricate than those customarily devised for controlling the insect pests of agriculture and are usually deployed on a state, national, continental or even global basis under the aegis of local or national health services or WHO."[23] Pest management in the field of public health has a number of special features that must be taken into consideration such as the urgency of dealing with an epidemic or highly endemic situations, and the need for simplicity and economy commensurate with the resources of the developing countries. Thus integrated pest management, by means of which several different control methods are employed to control vector populations in a particular ecological situation is likely to produce more effective and lasting results. However, the progress in this direction has been rather slow. Permanent results cannot be achieved unless detailed ecological studies have been conducted. Unfortunately, ecological approaches are not always appreciated owing to the fact that the advantages are not immediately felt. In countries where the necessary national expertise is not available, improved training facilities will be required to promote multidisciplinary team effort orientated towards the solution of practical problems.

Research efforts in this direction have been carried out either in the developed countries or under the sponsorship of international organizations in order to ensure the best expertise for developing suitable strategies aimed at the control of a few extremely important pests and vectors. Furthermore, the road to success is paved with many failures from which lessons can be learned to improve the sectoral approaches and overall management strategies including intercountry and regional cooperation. It must, however, be stressed that integrated pest management does not constitute a magic tool but is based on the sound and judicious application of the various control approaches that are available at a particular place and time.

DISSCUSSION.--The different methods for vector control described above should be taken into consideration in finding solutions to the problems of vector resistance and the present high cost and shortage of insecticides, etc. Although many of these methods are still not fully practicable, it might be expected that some of the projects involving them will show benefits. However, because of the complicated nature of the problem of vector control and the difficulties involved in integrating the different approaches, it will be necessary to retain the present proven methods despite their high costs and other disadvantages, especially when it is a question of dealing with emergencies.

Certain trends have emerged during the past few years which may be pointers to the future. These are briefly summarized below:

(1) In the field of public health, what is important is disease control and not vector control **per se**; thus, besides vector control, disease control must be carried out by the administration of drugs and vaccines, if available.

(2) The primary objective of the control program must be to reduce the vector population to a level below the critical density for the transmission of the disease, rather than to eradicate the vector completely.

(3) If long-lasting vector control is the objective, then permanent management of the breeding habitats must be provided for, even though this may require the outlay of capital which the developing countries may not be able to afford and so they must depend on external assistance.

(4) However, community participation, if efficiently mobilized, could be made responsible for relatively simple measures such as the maintenance and clearing of drains, control of vector habitats, spraying of insecticides, administration of drugs, etc. Reference may be made here to the patriotic mass movement in China for the control of four pests.[25]

(5) In emergency situations, provison must be made for the application of insecticides by ULV in order to control epidemics and possibly certain endemic conditions.

(6) Every effort should be made to strengthen emergency operations in addition to routine vector control activities.

(7) There is a great dearth of trained personnel in the countries where the problem is most acute. Therefore every effort must be made to ensure training and to promote self-reliance in these countries.

It is to be hoped that, in future, efforts will be made to integrate all available control methods - chemical, biological, genetic, and environmental, whenever feasible - in order to achieve maximum control at minimum cost. In this way, it is hoped that an increasing human population will live safely in the midst of vector populations that have been reduced to an innocuous level. Emphasis must be laid on the development of integrated control strategies which are cost-effective, socioeconomically acceptable, safe to the environment, and amenable to community action at the village level through primary health care. However, Corbet (1981) has recently summed up the situation that integrated pest management in the agricultural field is being impeded, especially in developed countries, owing to the continuing high level of pesticide use and the prevailing attitudes, i.e., behavioral and cultural traits.[26] It is hoped that the workers in the public health field will be able to benefit from the experiences of others.

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INTEGRATED CONTROL AND ITS APPLICATION TO ANTI-MALARIA PROGRAMS

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INTRODUCTION.–Integrated vector control as currently defined rarely has been planned and carried out in vector/disease control programs. Instead, reliance for control has been placed on a single method of vector control which appeared most cost-effective and feasible for application. Neither cost-effectiveness nor feasibility of application - two major criteria for selection of methods of control - were carefully assessed in and compared with other available methods. Likewise, other selection criteria, e.g. cost-benefit, impact on environment and long-term projection of costs, benefits, effectiveness, feasibility, etc., were not assessed for alternative methods and in other areas.

The malaria eradication program and the revised anti-malaria programs that have been planned and implemented follow the same trend. For vector control, almost total reliance has been placed on residual insecticides; other methods of control were not given consideration unless and until the former failed to produce a significant effect. One may acknowledge that the anti-malaria program was launched with haste and based on almost total confidence in DDT and its outstanding effects on mosquito vectors. The planners also believed that any delay in launching the program could deprive humanity of the beneficial effect of DDT insecticide, by then already widely used in vast agricultural areas.

The initial confidence in DDT and its impact was so pronounced that no consideration was given to the use of other measures and no attempts were made to assess their potential. This confidence was so complete that DDT residual spraying was universally advocated and applied, with the same dosage and often the same application cycle, irrespective of local conditions. Few programs changed the set standard or used lower dosages and other cycles.

The residual spray approach was effective in a wide range of situations, often despite operational imperfections. The insecticide was so potent and versatile that it was employed in many ecological and socio-economic conditions, with nearly universal success. Subsequently, a few years after initiation of the program, situations arose which required that this single vector control method be supplemented by others.

Numerous programs sought solutions through chemotherapy, combined with residual spraying. The primary problems related to exophylic vectors, including behavioristic avoidance of sprayed surfaces by vectors due to irritability or repellency qualities of DDT, and limited vector resistance. Operational difficulties of varying degrees were present in all programs, and problems of accessibility, communication, supervision and logistics continued to hamper efficiency.

With time, technical problems became greater; resistance developed in more vector species throughout increasingly larger areas, demanding more expensive and more toxic replacement insecticides, greater financial resources, and more complex and costly operational organizations.

The revised malaria control strategy adopted in 1969 began a more integrated approach. Other methods were reviewed, operations manuals were prepared, and staff training was reoriented toward integrated control. Achievements in the 13 years since malaria strategy was revised have been meagre and the progress toward applied integrated control strategies has been disappointingly slow. Programs still rely heavily on insecticides, particularly on residual compounds; unfortunately, the number of effective insecticides available for application against malaria vectors is rapidly decreasing due to increasing vector resistance, and the development of few new compounds.

There is general agreement that insecticides are now and will continue to be primary tools of anti-malaria programs for the forseeable future, and regulation and rational use is essential within the context of integrated control. "We must get away from a complacent insecticide program when we know that water and land management offer a more nearly permanent solution. We must get away from the philosophy that our job is to spray away the mosquitoes . . . . Rather we must think about mosquito control services and their total management and total IPM approach".[1]

METHODS OF VECTOR CONTROL; PRESENT STATUS AND USE.–Residual adulticiding is the most widely employed method of control in anti-malaria programs. Larviciding and space spraying have only limited use. The use of DDT in residual spraying has been declining steadily due to vector resistance. In most cases it has been replaced by malathion, the use of which has increased considerably in recent years. Fenitrothion and propoxur generally are used where there is resistance to malathion.

Table 1 shows that pesticides applied in larviciding and space spraying amount to only a small fraction of the amount of residual pesticides used, less than 0.05%. Only a few countries use larvicides and even fewer apply fog or ULV adulticides. There is little likelihood of significant extension of these methods in anti-malaria programs for the near future. Near total reliance on residual pesticides means that danger lies ahead in anti-malaria programs.

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[1] Proceedings of the Thirty-second Annual Meeting of the Utah Mosquito Abatement Association, 1979.

Table 1.--Possibilities for a comprehensive approach in operations against parasitic and other mosquito-borne diseases.

Disease	Principal vectors	Larviciding		Residual spraying		Space spraying & ULV		Source reduction			Biological		Sanitation		
		Chemical	Appli- cation	Chemical	Appli- cation	Chemical	Application	Filling	Drainage	Maintenance (corrective operations)	Fish	Environmental manipulation	Housing improvement	Water supply	Excreta disposal
Malaria	Anopheles	x	x	x	x	x	x	xⓧ	xⓧ	xⓧ	xⓧ	ⓧ	ⓧ	-	-
Filariasis	Anopheles Culicine	x	x	x	x	x	x	xⓧ	xⓧ	xⓧ	ⓧx	ⓧ	ⓧ	-	-
Dengue	Aedes	x	x	x	x	x	x	xⓧ	xⓧ	xⓧ	xⓧ	ⓧ	ⓧ	-	-
Yellow fever	Aedes	x	x	x	x	x	x	xⓧ	xⓧ	xⓧ	xⓧ	ⓧ	ⓧ	ⓧ	-
Encephal.	Culicine Aedes	x	x	x	x	x	x	xⓧ	xⓧ	xⓧ	xⓧ	ⓧ	-	-	-
Leish.	Flebotom. flies	-	-	x	x	x	ⓧ	-	-	-	-	-	ⓧ	-	-
American Trypanos. Chagas'	Triatoma bugs	-	-	x	x	ⓧ	ⓧ	-	-	-	-	-	x	-	-
Onchocerc.	Simulium fly	x	xⓧ	-	-	-	ⓧ	-	-	xⓧ	-	x	-	-	-
Trypanos.	Tsetse fly	-	-	x	ⓧ	x	ⓧ	-	-	-	-	-	-	-	-
Schisto.	Snails	ⓧ	x	-	-	-	-	xⓧ	xⓧ	xⓧ	-	ⓧ	-	x	xⓧ
Protozoa and helminthic intestinal infections (soil-borne infections)		-	-	-	-	-	-	-	ⓧ	-	-	-	ⓧ	x	x

x = Greatly - (great possibility exists that the same chemicals, methods and equipment will be effective against all diseases marked with an x).

ⓧ = Partially - (the possibility exists that the same chemicals, methods and equipment will be partially effective or on certain occasions against diseases marked with ⓧ).

- = Not at all - (little or no possibility exists that the same chemicals, methods and equipment will be effective against other diseases marked with -).

Except for larvivorous fish, biological agents in anti-malaria programs are mainly limited to field trials. Use of the mosquito fish, *Gambusia* sp., was begun in the last 1960s in the Middle East, and within a few years a number of programs successfully employed this fish in extensive anti-malaria programs. The use of native fish for mosquito larval control has received special attention, and some countries have used the guppy, *Lebistes reticulatus*. *Tilapia* sp. have been used successfully in rain water collection cisterns, etc. Mature *Tilapia* are a source of food for the local human population when the water disappears in the dry season.

Environmental management rarely has been used. Simple drainage and filling has been employed in a few programs to a limited extent. Promotional steps have been initiated and the extended application of these measures in health programs and in agricultural water resources development projects is being actively pursued. Training aids and institutions, field trials and studies, as well as interdisciplinary collaboration vital for successful IPM operations are being organized.

INTEGRATED CONTROL OR IPM*.–In simple language, integrated control can be defined as "the selection and application of methods of control to optimize achievement of results." This implies that all methods of control must be considered and assessed but only those that produce practical results will be used singly or in combination.

Combining methods in a control strategy is not always necessary, but may be a means to optimize achievement. Thus, programs may depend on only a single method of vector control when investigations prove other methods locally unsuitable or ineffective or unjustifiably costly in relation to the results to be achieved. The optimization may range from more effective control of a vector or of the disease(s) it transmits, to better protection of environmental quality, improvement of economics of operation, securing long-term benefits, increasing correlated benefits, general health and socio-economic improvement. The priority and values attached to each factor may vary in each situation or program and thus integrated control may not necessarily be always more effective or economical or less harmful to the environment than the conventional approaches. Priority may be given to one or more factors, depending on the program objectives, priority problems, resources available and the local social, cultural and political conditions. The integrated control program must be designed to achieve the objectives following the established standards and criteria. The program objectives and priority targets must be clearly defined to enable the design of integrated strategies that best meet established priorities.

APPLICATION OF INTEGRATED CONTROL TO HEALTH PROGRAMS.- Integrated control may prove complex to organize. The three components as defined, namely selection of methods of control, their application, and optimization of results, may be considered in the complex task of identifying the related "systems" and numerous "elements", their interactions, and in studying and assessing the several alternatives arising from their combination and integration. The systems involved may include environmental social, political and cultural, and major elements generally include vectors, parasites, and man and his environment. In every environment and social situation, different sets of interactions occur. Thus, with our inadequately precise methods of survey and measurement and the paucity of valid standards, it may prove impractical to select from among the multitude of possible combinations the best set, or the strategy that fully optimizes the results. Until more precise data and standards are available, one may need to rely on improvization for assessment and selection, and for optimization in achievement of results.

Integrated control in vector/disease control and IPM in agriculture or in mosquito pest management have the same basic principles, though differently defined, or different in composition and structure, and directed towards different objectives. For example, IPM in agriculture includes the three major elements of pest, plant, and environment; integrated control in health also involves a fourth element, namely man, thus further complicating an already complicated process. In agricultural pest management, the assessment of economic losses or the setting of an economic threshold as an essential part of the pest management planning process can be done with a fair degree of accuracy. In vector management, the process is much more complicated and generally the results are only approximations or arbitrary. Measurements, of benefits/ cost of health projects, and sometimes measurements of the effectiveness/ cost have proved impracticable. Similarly, it is more difficult to identify the elements involved in vector management and their interactions and to assess their roles and impacts. The establishment of an acceptable economic and social threshold or injury level for a disease is much more difficult or may be unrealistic. Much research and field investigation will be required and more extensive use may have to be made of mathematical modeling and simulations to identify the elements and their interactions, to analyse and predict the possible impacts, and to propose appropriate feasible strategies.

INTEGRATED CONTROL IN ANTI-MALARIA PROGRAMS

The application of an integrated approach to anti-malaria programs required the adoption of a number of principles and practices and their planning and gradual integration into each program. Although the principles of integrated control may remain the same in every program, the practices of this approach and the planning of the activities may vary from program to program depending on local conditions, program objectives, and resources available.

Applying the principles of integrated control relates to the three components of the definition (namely selection and application of methods and optimization of results) to anti-malaria programs. The program objectives should be defined in order of priority. It is necessary at first to decide whether the program will be an eradication or a control program. If the latter is chosen, now usually the case, it is necessary to determine what level of control is desired and how soon this must be achieved. Then the available methods of control should be reviewed and those considered feasible for applica-

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*Integrated Pest Management

tion under local conditions should be assessed for effectiveness/cost, benefit/cost and benefit/risk, on both short and long-term bases. Conditions normally vary from area to area and therefore the assessment may need to be preceded by a stratification of operational areas according to ecological variations and perhaps also on a social, cultural, economic, and political basis.

The practical follow-up and implementation of the steps stated above often is a complex task. Due to the inadequately precise methods of measurement and the paucity of valid standards in current use, the true assessment of conditions and thus the proper selection of methods of control and their effective integration will be difficult. Nevertheless, surveys and field investigations must identify ecological conditions influencing the prevalence of the vector(s), its role in disease(s) transmission, the disease epidemiology, and the socio-economic factors affecting the system. Field trials may have to be organized in several ecologically and socio-economically stratified areas to measure the impact of various methods of control and the feasibility of application in each stratum.

Measurement of effectiveness/cost can be based on the degree of reduction achieved in vector population density, or of the specific group against which the control measure is directed, and on total cost of the anti-vector control measure. The measurement of effectiveness may be based on the impact of the measure or of the strategy on the disease(s). In this case, other factors may intervene and thus the results cannot be related directly to vector control measures alone. Effects and costs must be projected over a long period of time and calculated for each measure before they are analysed and compared for selection. In this projection, the beneficial and adverse effects of a method on other health problems and in other social and economic fields, e.g. agriculture, urbanization, and tourism, should be assessed and included.

An assessment of benefits may be more simple if related to tangible benefits, e.g. those arising from land reclamation, agricultural improvement and provision of extra water for irrigation or consumption, or better housing, etc. It may be difficult to evaluate intangible benefits in money value, e.g. health improvement leading to increased productivity, reduction in suffering and pain due to disease and the lower cost of the health care system. Several unsuccessful earlier attempts were made to quantify these levels, although their positive impacts are universally accepted. The assessment and quantification of benefits/risks is equally complex and therefore the assessment process should be based more on identification and study of possible adverse effects in formulating integrated strategies, rather than on their quantification. Research and field investigations are underway and should be intensified to produce more precise methods and standards for identification and better assessment of effectiveness, benefits and risks.

PRACTICAL APPROACH TO IMPLEMENTATION.–From the preceding, it may be apparent that integrated control will be the optimum approach to vector control, but its practical application will be complex and time consuming. There is an obvious need for more information and data on vector ecology, disease epidemiology, and related environmental and socio-economic conditions. These data form the basis for stratification of areas and for consideration of various methods of control in each stratum. Collection of such data and their compilation and analysis require highly qualified staff in the professional category, and training of intermediary and field staff. The application of integrated control can be attempted in two steps that may be concurrent:

Pilot operation. A practical approach would be to select at the initial stage a pilot operation area in one of the regions currently served by anti-malaria operations. The size of the area will depend on the resources available but should have a population of at least 100,000 in rural areas to allow reliable epidemiological evaluation. A qualified professional staff (epidemiologist, entomologist, sanitary engineer, etc.) should be assigned the task of training local personnel of the area in the collection and compilation of data. When the data are analysed, the project can be designed, including methods of control to be selected and tested, the integrated control strategies to be formulated and assessed, application of the selected strategy and evaluation of the result. The area can then serve as a training project for other staff, and as a central point for progressive extension of the integrated approach to other operational areas.

Improvement and reorientation of current operations. Concurrently with selection and operation of the pilot area, the methods of control then in use should be reviewed and adjusted to better comply with the principles of integrated control, as well as studying possibilities for introduction or extension of other available methods that are not used or are applied on a limited scale.

Most programs are emphasizing wide-scale use of residual spraying; accordingly, the review may begin with assessment, correction and improvement of this method, aiming primarily at counteracting vector resistance. The following steps are proposed:

(a) Review the geographical reconnaissance and mapping for adequacy and exactitude, including up-dating.

(b) Review the state of ecological, social, economic and environmental information available, and determine if these data are used in actual planning and implementation of the program.

(c) Examine the suitability of the pesticides and formulations used in relation to the expected residual effect, type of surfaces to be sprayed, safety to man and environment, compliance with specifications, local suitability, shipping, storage and distribution conditions.

(d) Review field operations, the training of staff, equipment for application, the techniques of spraying, coverage in time and space, accessibility of villages, communication systems, housing, population reactions, transport, logistics, etc.

(e) Check that timing, dosage, cycles and coverage of application relate closely to the local seasons of prevalence of vector species, the disease transmission season(s) and the length of residual effect expected from the pesticide used. The period of residual effect is also related to the objectives of the program and the degree of control required.

(f) Determine that the supervision and evaluation systems are adequate and regular and that the results are used to correct and re-adapt or reschedule the control strategy. The evaluation should include an assessment of costs and their analyses with respect to various cost centres.

(g) Determine whether the service is properly organized and conveniently placed within the Ministry of Health structure, with functional links to facilitate collaboration with other related health departments, ministries and agencies, e.g. agriculture, irrigation, and public works, etc.

When the program review is completed, the following steps may be taken toward an integrated strategy.

Rationalize the use of residual pesticides. This function may include:

(a) A study of the possibilities of gradually replacing residual spraying in certain situations, e.g. hypoendemic areas, arid areas, by integrated control using other methods of chemical control, biological and environmental management measures, and chemotherapy.

(b) An examination of the possibility of supplementing residual spraying in some areas by other methods to reduce the intensity of spraying, e.g. coverage, dosage and number of cycles of application.

(c) A more precise adjustment of the timing, dosage, cycle, and coverage of spraying to satisfy local ecological conditions and to insure that these factors lead to achievement of the results aimed at. In many anti-malaria programs aiming at control, the spraying operation is sometimes applied unnecessarily, to interrupt transmission, e.g. in areas where the malariogenic potential if reduced to a very low level cannot be maintained. In these areas the intensity of the residual spraying can be reduced to prolong the service life of the insecticide and to save resources.

(d) Extension and intensification of training of staff, especially that of field supervisors and spraymen, to apply insecticide more efficiently as prescribed for each situation and with care to prevent avoidable human and environmental contamination.

(e) Improvement of logistics, transport and equipment used in field operations, to facilitate timely application and efficient supervision.

(f) Collaboration with the Ministry of Agriculture to limit and if possible avoid the use of public health residual pesticides in agricultrue to delay the development of resistance by local vectors.

Application of chemical larvicides. Larvicides are currently applied in anti-malaria programs mostly in urban situations and usually combined with other methods of control, e.g. chemotherapy, space spraying. Application of larvicides in rural situations has been limited mainly due to operational complexity and higher cost per capita, especially in areas where water is abundant or rains frequent. Under these conditions identification of breeding sites, application of correct dosage of pesticides, and maintenance of adequate dosage have been difficult. In arid and semi-arid areas effective larviciding in rural areas may be more feasible but field supervision has always been a problem, and for effective results, operators must be thoroughly trained and conscientious.

Nevertheless larvicides must be used increasingly as a supplementary measure wherever conditions allow, in order to alleviate dependence on residual pesticides. Effective and relatively safe pesticides of non-residual groups are available. More efficient equipment is available and can be used to extend the use of larvicides and increase their efficiency.

Space spraying of pesticides. Application of pesticides in aerosol form for adult vector control has not been given the importance it deserves. With difficulties encountered with residual spraying in anti-malaria programs, ranging from resistance of the human population to staining of painted houses, to odour of certain compounds, and the short residual effect of most replacement residual pesticides as well as the limited number available for operational application, the application of non-residual pesticides in aerosol form should be seriously reconsidered and extended. When combined with other methods of vector control and chemotherapy space adulticiding can produce effective malaria control.

Aerosol application can be done indoors and outdoors to effectively control both domestic and peri-domestic mosquito populations. Effective insecticides in non-residual groups can be applied as cold or thermal fogs or ULV. Also, high performance versatile applicators, hand and back-carried or vehicle mounted, are available for use in rural and urban areas. Aircraft are frequently used in large scale operations, and with availability and proper maintenance facilities, they can be used in larger anti-malaria programs to reduce application costs.

A major problem with aerosol application of pesticides is the need for frequently repeated operations. Cycles to two weeks to a month may be necessary for effective control. Taking into consideration the three monthly cycles presently needed with the replacement pesticides in residual spraying, and the much greater speed of application of aerosols, and their wider effectiveness extended to peridomestic resting places, in many situations the use of this method may be favoured and be appreciably cost-effective. Because application operations can be done speedily, the field organization and staffing of aerosol application operations will be considerably simpler and fewer staff will be required.

Aerosol application of pesticides will be an effective component of integrated control and should be considered when designing integrated strategies.

Use of biological agents. Of the biological control agents, only the larvivorous fish are commonly employed in operational applications. *Gambusia* sp. has been used in many large scale programs with good results. Other larvivorous fish, e.g. *(Lebistes reticulatus)*, guppy and *Tilapia* sp. have also been used operationally.

To be effective, the fish population should be adequately high. Experience in Asian countries indicates that a density of 5-6 fish per square metre is necessary for effective mosquito control. In tropical areas the fish may thrive all year round and maintain adequate density. Conversely, in areas with cold winters the fish may die in shallow breeding areas during the cold months and the population has to be replenished in the spring. Production of a significantly great number of fish within a short period has been an operational problem.

Larvivorous fish are used in a wide variety of breeding places, including ponds, cisterns, wells, irrigation and drainage canals, and rice fields. Dense vegetation lowers the efficiency of fish, and concurrent use of herbivorous fish is being studied and should prove useful.

Introduction of larvivorous fish for vector control is a simple, inexpensive, and reasonably effective measure of vector control, and should be considered as a component of integrated strategies. Use of fish is especially indicated in combination with environmental management for controlling limited residual breeding in reclaimed areas. In principle, use of native species should be given preference to avoid possible introduction of undesirable new species.

Environmental Management. Of all methods of vector control, environmental management measures have been least used in anti-malaria programs. However, these were the measures first used in the early part of the 20th century to control malaria in many parts of the world. Neglect of the environmental measures is due perhaps more to the lack of awareness of these measures and their values and effect than to inability to plan and implement them. Steps have been taken in recent years to promote environmental management for vector control and to train staff in related techniques.

By reason of long-term beneficial effects, environmental management methods may be more cost-effective in many situations than other methods of vector control and should be an essential component of an integrated strategy. Simple and low cost source reduction measures should be introduced into anti-malaria programs, while more sophisticated methods need to be integrated into development projects. In these projects, integation of environmental management methods should be considered from the initial stages of project planning and included in the design, construction, operation and maintenance of the project.

Promotion and implementation of integrated control in anti-malaria programs. Promotion and implementation of the integrated approach obviously must start with information and training of senior staff at decision-making level. It is essential that program directors and senior staff, as well as the policy makers at higher levels in the Ministry of Health, recognize the advantages and requirements of IPM and be convinced of the need for it. An integrated approach and its successful application depends largely on the efficacy of collaboration with other related ministries and agencies. Institutional arrangements for this purpose need to be established at national and international levels. The following steps have already been taken:

(a) A seminar on integrated control of mosquito vectors for the senior staff of the anti-malaria services and the WHO advisory teams has been planned and will take place in Turkey, November 2-14, 1981.

(b) A joint WHO/FAO/UNEP Panel of Experts on environmental management for vector control has been organized to promote collaboration at national and international levels for the promotion of these activities for vector control. (First meeting: September 22-29, 1981).

(c) Pilot operations have been proposed to be organized in anti-malaria programs in each WHO region to train staff and to promote the application of simple environmental management measures in anti-malaria programs as a part of integrated strategies.

(d) A WHO comprehensive manual on environmental management measures for vector control, emphasizing integrated control, has been completed and is in print. A similar manual on chemical control is being completed.

(e) The first expert committee on environmental management for vector control was held in November 1979.

Comprehensive approach to vector/disease control. The scope of integrated control in anti-malaria programs can be broadened to include other diseases that may be affected by the program.

Experience has shown that as a result of operation of an anti-malaria program with indoor residual spraying, the incidence of some other diseases such as leishmaniasis or Chagas' disease has been reduced, sometimes significantly. Other methods of mosquito control used in anti-malaria programs have similar effects on vectors of other diseases. In some instances, a little modification in the techniques or practices of application or the material used may render application of one method effective against a number of vectors and diseases.

Similarly, pest control in agriculture and development of water resources through the construction of dams, irrigation schemes or land reclamation may have both positive and negative effects on vectors and diseases they transmit. Minor modification or adjustment of techniques, design, and material may produce significant assistance to vector/disease prevention and control.

It is obvious that a broadening of the scope of integrated control into a comprehensive approach involving multi-sectorial collaboration will produce significant gains in vector/disease control and this perhaps at little or no extra cost. In today's tight economic situation and acute shortage of funds for health promotion and development, the comprehensive approach seems to contain realistic promises and prospects for success.

This approach is now being tested in a large-scale project - The Blue Nile Health Project in the Sudan. The project covers the Gezira/Managil and Rahad irrigation schemes with 2.2 million acres of irrigated land and 2 million transient and settled population. Malaria, schistosomiasis, and diarrhoeal diseases are being tackled in one comprehensive strategy involving other health services and agriculture and irrigation services, as well as local educational and socio-cultural institutions.

The project began operations in April 1979, and is expected to be completed in 10 years at a cost of $150 million. It will be used as a training ground for staff from various related sectors in preparing for development of comprehensive strategies elsewhere.

A World Bank Rehabilitation and Modernization project (US $150,000) is being planned for the Gezira Irrigation Scheme. It includes activities which will have considerable beneficial impacts on the present disease/vector situation. In addition, the project will include a health component contributing directly to certain operations of the **Blue Nile** Health project, e.g. water supply, environmental health management, etc.

MOSQUITO CONTROL IN THE UNITED STATES AIR FORCE DURING THE 1980'S[1]

Dennis D. Pinkovsky[2]

ABSTRACT

Results of a questionnaire on mosquito control sent to 135 U.S. Air Force (USAF) bases are summarized. Of 55 responding installations, 52 had mosquito control programs: 11.5% used biological control methods; 61.5%, physical mosquito control means; 67.3%, chemical larvicides; and 90.4%, chemical adulticiding measures. Malathion emulsifiable concentrate for larval mosquito control and ground ultra-low-volume applications of technical grade malathion for adult mosquito control were the primary chemical approaches used.

INTRODUCTION AND PROCEDURES. USAF personnel have used integrated tactics to control mosquitoes on their installations for many years. A survey was made in late 1981 to determine specific techniques, and relative importance of those techniques, used during the previous two summers for mosquito abatement in the USAF. The Medical Entomology Section, USAF School of Aerospace Medicine, Epidemiology Division, Brooks AFB, Texas, sent a questionnaire to Civil Engineering Entomology Sections at 135 bases in the continental United States (CONUS) and overseas requesting: 1.) an indication of the biological, chemical larviciding, chemical adulticiding, and physical control methods used to control mosquitoes, and 2.) the percentage of each installation's total control effort that was expended on each technique. Pest management professionals at major air command headquarters also provided summaries of overall mosquito control activities in their commands for individual fiscal years.

RESULTS. **General.** One overseas base and 54 CONUS installations responded (these bases represented seven major air commands and 31 States.) Of the responding installations, the overseas base (Sembach AB, Germany) and two CONUS bases indicated that they performed no mosquito control; 52 of the CONUS bases (96% of total respondents) indicated that they accomplished some mosquito control. Six bases (11.5% of positive respondents) used biological control methods; 32 (61.5%) used physical techniques; 35 (67.3%) used chemical larvicides; and 47(90.4%) used chemical adulticiding measures.

Biological Control. Larvivorous fish (*Gambusia* and others) were actively stocked at five bases and insectivorous birds (purple martins) were used at one. A few bases have experimented with the bacterial mosquito larvicide *Bacillus thuringiensis* var. *israeliensis* (BTI); however, no installations listed BTI as part of their routine mosquito control program. BTI will probably be used more as its effectiveness and environmental soundness are better recognized, as additional formulations become available, and as increased usage lowers the material's cost or when BTI is Federally stock listed.

Physical Control. The physical control methods (used by 32 of the bases) included artificial-container removal, ditching, draining, insect electrocution traps, filling, and vegetation removal. The latter two measures--filling (including relandscaping) and vegetation removal (including use of burning and aquatic herbicides)--were each used at 16 bases. Draining and ditching (reditching) were used at 14 and 13 bases, respectively, and one base each indicated regular use of insect electrocution traps and removal of discarded artificial con-

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[1] The views expressed are those solely of the author and do not necessarily represent the opinions of the US Air Force or the Department of Defense. Mention of a pesticide or proprietary product does not constitute a recommendation or endorsement by the author or the US Air Force.

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tainers to limit mosquito populations. Mosquito control in many instances is a secondary benefit of physical activities such as vegetation control or reditching that are done primarily for aesthetic reasons or to avoid flooded conditions.

Chemical Larviciding. Seven different products were used in chemical larviciding programs for mosquito control. Malathion emulsifiable concentrate was used most frequently and accounted for an average of 29.4% of the total control efforts of the 16 bases using it (range 5-90%). Malathion used as a larvicide could hasten the development of insecticide resistance since 15 of the 16 installations also used malathion for adulticiding. Dursban® was used at 13 (25%) of the bases with mosquito control programs and accounted for an average of 15.3% of these bases' total mosquito control efforts. Dursban® 10CR granules were used at 11 bases and the emulsifiable concentrate was used at two. Malathion and Dursban® are Federally stock-listed items. Flit MLO or light mineral oil was used at eight (15.4%) of the bases, and diesel fuel at seven (13.5%). Fewer than 10% of the bases used pyrethrum Tossits® (7.7%), Abate® (5.8%), or Altosid® (3.8%).

Chemical Adulticiding. Chemical adulticiding activities included ground ultra-low-volume (ULV), thermal and cold fogger, Buffalo turbine, and aerial spray applications and involved four insecticides: Dibrom®, fenthion, malathion, and pyrethroids. Of the 52 bases, 39 (75%) used ground ULV delivery of malathion. ULV malathion applications accounted for an average of 66% of the total mosquito control efforts at these bases (range 5-100%). Only three bases used Dibrom® in their ground ULV machines, and one base applied ULV fenthion. Thermal foggers were used to apply malathion at three bases and pyrethrins at one. Cold foggers were used at two installations to disperse malathion, and one base each used a Buffalo turbine mister or applied residual malathion for adult mosquito control. Aerial applications of Dibrom® were used to kill adult mosquitoes at three installations; the USAF Reserve Aerial Spray Branch did the air sprays at two of the bases, and the local county performed the air sprays at one. Malathion was applied by air spray at one base. Malathion or other insecticide resistance in mosquitoes is not a recognized problem at most bases; however, field testing is called for to confirm the continued susceptibilities.

Commandwide Summaries. Estimates of acreages treated in mosquito larviciding and adulticiding efforts for various major air commands during FY 81 (October 1980-September 1981) ranged from 38,084 acres in Air Force Logistics Command (which controls six bases) to 1,329,425 acres for Tactical Air Command (which encompasses 17 installations). For five reporting air commands during FY 81, a total of 2,194,017 acres were treated and 30,810 manhours and an estimated $385,235 (materials and labor) were expended on mosquito control.

CONCLUSION.–The preceding comments illustrate that USAF installations use a varied approach to mosquito control with continued emphasis on chemical control and substantial physical control efforts.

BLACK FLY CONTROL IN SALT LAKE COUNTY

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Black flies have been abundant in Salt Lake County for many years, (Rees and Peterson, 1953). For some reason, not yet known, they became a severe nuisance beginning in 1967 with the nuisance being more severe in some years than in others. The flies did not bite man in the Salt Lake Valley but swarmed around the head in such numbers as to make outdoor activities extremely uncomfortable. Peterson (1956) also noted that the flies in Salt Lake County do not bite man below 7000 feet. Pressure from the public mounted on the South Salt Lake County Mosquito Abatement District to control the pest and in 1974 the Board of Trustees of the District ordered studies to begin with the ultimate goal of abating the nuisance.

Studies were started by the District in 1975 and a small pilot control program was started in 1978. The program has been expanded since and funding will be available for a full scale program in 1982. This slow development was due both to funding and because the District wanted basic information on the black fly biology, life history and ecology including other organisms in its aquatic environment.

Black flies have been in the Valley for years but only recently have complaints been phoned into the District office. In 1980 over 50 complaints for gnats were received and less than 10 for mosquitoes in the same time period. Inordinate numbers of black flies the past few years have rendered sporting and recreational areas such as golf courses, ball parks and picnic facilities less than enjoyable due to harassment from large numbers of flies swirling around the head area. Outdoor activities are severely hampered everywhere in the Valley during August and September. Reports from animal owners have indicated severe damage to ears of horses and dogs. Peterson (1956) has some questions about specific host preference but does list man, horses, cattle, turkeys and ducks as some of the animals known to be fed upon.

The capacity of the black fly to disperse over great distances in any direction [Fredeen 1956 (1958), Stoltz 1981] provides little escape from the pest anywhere in the Valley. With a human population of about 650,000 and many animals, both farm and pet, the nuisance to both is obvious as well as is the potential existing to do physical damage to livestock.

After limited surveys were conducted in 1975 to determine where and at what densities black flies occurred, the District Board ordered a full fledged survey to determine distribution, density, and the ecology of the fly and examine the possibility of control. Coupled with this order was the need to sample the respective streams and River for existing populations of "nontarget" organisms.

Early Utah studies (Knowlton and Rowe 1934, Stains and Knowlton 1940, 1943) identified specific flies and their biology and location but little was known of the general distribution and numbers of black flies until Bobbie V. Peterson did his work during the 1950's (Peterson 1953, 1955, 1956, 1959, 1960). With his contribution much of the initial information of black fly biology, ecology, taxomony and distribution was provided. Rees and Peterson (1953) and Peterson (1960) report that black flies are found in virtually every stream at every elevation in the Salt Lake area. A survey by Peterson (1958) indicates *Simulium vittatum* Zetterstedt is the predominant inhabitant of the water ways in Salt Lake County. This species presents serious problems to the area because of its tendencies to utilize a wider variety of stream conditions and to disperse over great distances [Fredeen 1956 (1958), Peterson and Wolfe 1956 (1958)] with population densities increasing during the summer until intolerable levels are reached in late summer and fall.

All known running sources of water in the District were surveyed for black fly larvae. Even trickling ditches 6" wide were found to contain heavy concentrations of flies attached to exposed tree roots. Because of the flight ability of black flies even streams coming from nearby canyons, of which there are several, were checked. All producing areas were mapped and coded as well as those few thought capable of future production. Specific treatment sites were selected and numbered to provide consistent data as to downstream efficacy from any given point.

Four different types of water habitats for black flies were identified. These included a small river (Jordan) varying in depth and width as it flows North from the Utah Lake to the Great Salt Lake; several clear water canyon streams; irrigation laterals or ditches and irrigation canals. Four areas of the Jordan River were sampled, (Table I), two areas of a shallow (12-18" deep), clear nature and two areas with heavy silting. The first two sampling areas are below a diversion dam with rocky substrata and trailing water weed (Potamogeton). The other two sources included a rocky shoal along the edge of a bend in the River (12200 S.) and the rocks used as a diversion under a bridge (7800 S.). Samples were collected by removing all materials in a square foot sampler: rocks, vegetation, sand and water, followed by hand sorting and washing using various screens, then preserving the specimens in 70% alcohol until identification could be made. Big and Little Cottonwood Creeks were sampled in two areas with similar riffling locations

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TABLE I

SITES SAMPLED DATES/COLLECTED

MACRO ORGANISMS COLLECTED	15300 S. Jordan River (Below Dam) (9/19/79) Nos.	15250 S. Jordan River (Riffles) (9/20/79) Nos.	12150 S. Jordan River (Stays) (9/27/79) Nos.	7800 S. Jordan River (Bridge) (10/14/79) Nos.	Big Cottonwood Creek (Walker Lane) (10/3/79) Nos.	Little Cottonwood Creek (Red Barn) (9/28/79) Nos.
Trichoptera						
Hydropsyche sp.	449	76	687	2517	8	94
Ephemeroptera						
Callibaetis sp.	5		4			
Leptophlebis sp.			16			
Beatis sp.				10	12	
Diptera						
Simuliidae						
Simulium vittatum	1474	166	73	82	1	2
Chironomidae						
(Chironomis sp.)	66	116	42	86	5	18
Empididae						
(Hemerodromia sp.)	16					
Stratomyidae					1	
Rhagionidae		1		2		
Dolichopodidae		1				
Coleoptera						
Elmidae						
(Narpus sp.)	31					
Hemiptera						
Ambrysus sp.		17				
Hirudinea		106	19	1	1	3
Nematoda	3					
Odanata						
Hetaerina sp.			1			
Crustacae						
Gamerus sp.	44	382	27	1		
Molussca						
Gastropoda	5		4		26	1

that stayed wet more frequently than other stretches of the streams due to spring water inflow.

S. vittatum is the prevalent species in the Valley. It is wide spread in diversified habitats and the larval form which can overwinter tolerates high temperatures, low pH, low oxygen tension, sewage effluent and silt or other suspended materials. Other species reported in Salt Lake County (Peterson 1958) are: *Prosimulium exigens* Dyar & Shannon, *P. onychodactylum* Dyar & Shannon, *Eusimulium aureum* Fries, *E. canonicolum* Dyar & Shannon, *Simulium arcticum* Malloch, *S. argus* Williston, *S. hunteri* Malloch, *S. twinni* Stains and Knowlton and *S. venustum* Say. Identification of current collections of sources is still being undertaken.

As mentioned earlier, there are at least four major water types producing black flies in the County.

1. The Jordan River, a slow moving (one - four feet per second), polluted stream that bisects the Valley from South to North starting out of the County boundaries and ending out of the control district, emptying into the Great Salt Lake some 15 miles Northwest of the control area. The River moves as a sluggish body for most of its length with the exception of the Jordan Narrows, a bluff area cut through by the River at the South end of the County and here there are shallow rocky stretches with trailing aquatic vegetation as well as the vegetation found trailing in the water from side banks. The last 15-20 miles are deeper (three - five feet) with heavy loads of silt and organic material from incoming irrigation and sewage treatment effluent.
2. Bordering the Valley to the East is the Wasatch Mountain Front and to the West the Oquirrh Range. Several canyon streams flow across the Valley to the River. These range from small, trickling, temporary sources to two large, fairly constant streams, Big and Little Cottonwood Creeks. These streams are clear, have rocky bottoms and are shallow through most of the season. The large ones have periods of intermittent drying up as they are two sources of culinary and irrigation water. Larvae are found in large numbers on the rocks in the streams and on trailing vegetation during high water periods.
3. Irrigation ditches crisscross the temperate desert valley and are intermittent at best. However, several of the large ditches which are almost continuous in flow produce large numbers of larvae several times a year. These ditches are silt laden, move slowly (.5-1 f/s). They have areas of still water that makes effective treatment difficult downstream. Larvae have been found on rocks, on ditch bottoms and trailing vegetation.
4. Irrigation canals supply the major portion of water to the irrigators along the Valley floor. They run three-five feet deep, move slowly (1-2 f/s) and contain heavy concentrations of suspended matter. The trailing vegetation is thick along the banks and provides excellent attachment sites for black fly larvae.

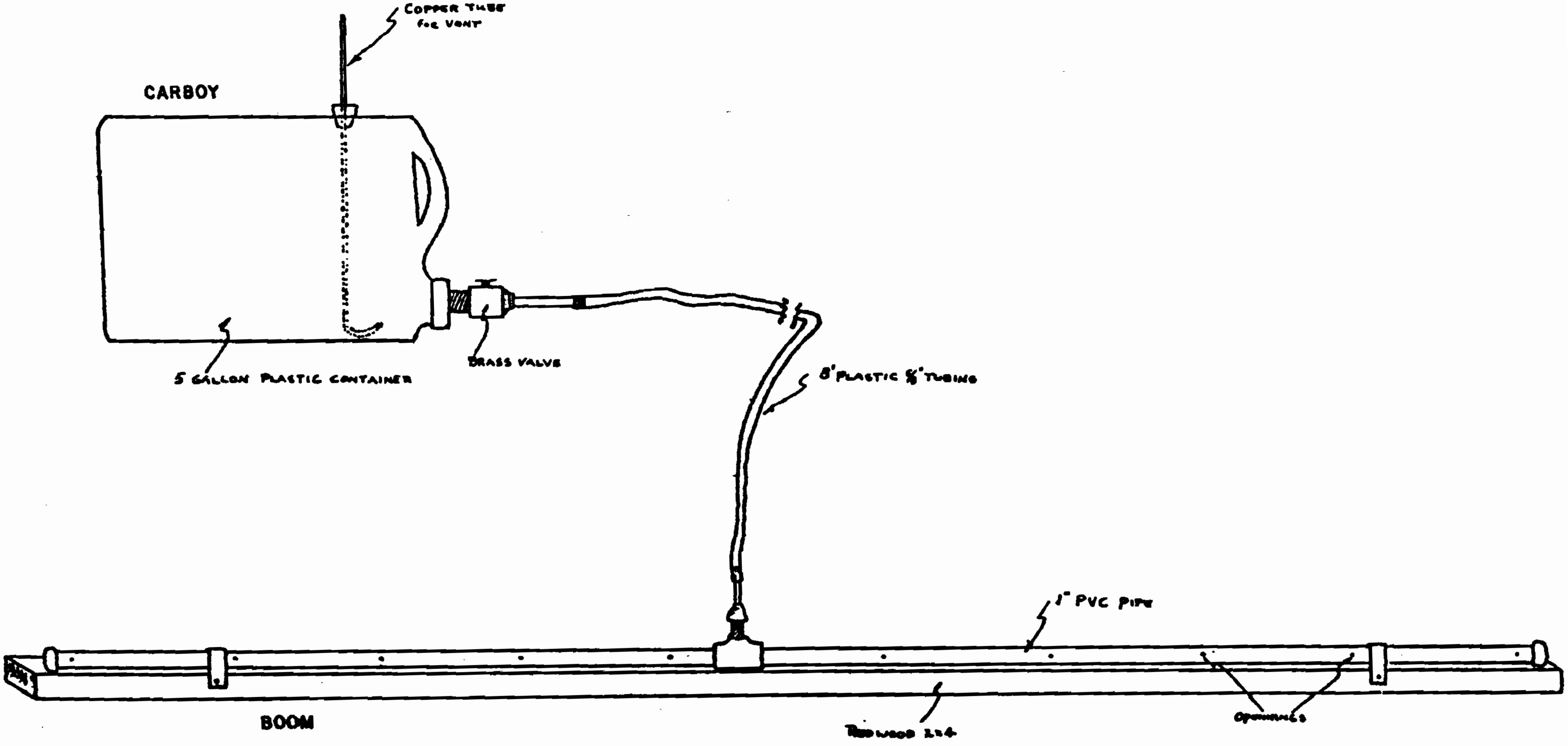
Each of these systems provides their own special problems when it comes to control. Slow moving, heavily silted waterways don't seem to provide the carry of the pesticide that fast, turbulent silted streams or rivers do such as the Athabasca waterway. Adsorption to suspended particulate matter appears advantageous (Haufe 1980, Charnetski et al., 1980) only if they remain in suspension. Through Salt Lake County the movement is so slow that suspended material doesn't remain suspended the length of the needed treatment area and efficacy for long distance treatment is still being tested. Dead water areas behind headgates, diversion dams along the River, larger canals and streams cause suspended matter to drop out and further complicate the problems of getting good control downstream from the point of treatment. Pesticides drop out of solution at these areas sometimes in sufficient quantities to reduce concentrations below effective strength. At present it is not practical to treat the Jordan River and canyon streams at their source and reinfestation from larval drift occurs as the insecticide is removed by stream flow. This obviously hampers efforts downstream in determining effective control at any given check point,(Waters 1965, 1969, 1970, Pearson and Franklin 1968). This problem has been noted by others.

In order to provide meaningful data, some type of standard measuring device must be used. There are many recommended (Gersabeck and Merritt 1974, Wolfe and Peterson 1958) but in an ongoing control program with so many different water problems, a standard device for all hasn't been developed. Tiles or bricks or similar devices put into our irrigation ditches silt over before they can be effective. Trailing plastic devices in the water where alternate drying exists doesn't work because the plastic becomes brittle and breaks to pieces in subsequent flooding. A device that seems most effective, at this point, in most streams is a yellow poly propylene 3/8" twisted rope. This device can be cut to length desired and placed in any stream along stream banks with a wire attachment or trailed from low bridges crossing streams. Markings on the rope using plastic electrical tape can delineate the areas desired for counting. Modeled after a device by Stoltz (1981).

Existing rocks in the streams that are marked previously or trailing bulrushes or cattails or tree branches can act as indicators of larval populations and control but no effort has been made to use these as quantitative data collecting devices for long range studies because of obvious inconsistencies.

Equipment used to dispense the pesticide is a modification of that described by Fredeen (1970). Two redwood booms were constructed to dispense materials into streams; one 5 feet long for narrow areas, and the other 10 feet long for larger streams, i.e., the Jordan River, Big and Little Cottonwood Creeks. On inch PVC pipe was attached to the redwood 2 X 4 and 1/8" holes were drilled one foot apart to allow the formulation to distribute evenly into the stream. This was attached to a five gallon plastic water jug modified to the carboy standards with a monitoring valve and plastic tubing (See Figure I). The booms were held in place in the stream using chains attached to eyes screwed into the ends of the boom. Where the streams and river have been too wide, the pesticide has been released through the tubing only directly into the water.

Figure I



BLACK FLY CONTROL APPARATUS

Many workers apparently have had the same problems with this kind of introduction method in getting the pesticide to dispense rapidly throughout the total medium to the desired toxic levels to minimize damage to non-target organisms (Haufe 1980). While putting the pesticide in at sites where the water is turbulent (dam spillways, headgates, etc.), complete mixing still takes several hundred yards before success is noticed all through the stream. We are working on a different method of application using a pump arrangement and will report on this at another time.

The insecticides considered for treatment in Salt Lake County were Abate (0,0,0′ , 0-tetramethyl 0,0′ - thiodi-p-phenylene phosphorothioate) and Methoxychlor (1,1,1-trichloro-2,2-bis(p-methoxyphenyl)ethane). These are proven insecticides against black fly larvae in Africa (WHO/VBC/76.653 Anon, Stiles and Quelennec), Canada, (Helson and West 1978, Baldwin et al. 1977, Wallace et al. 1973) and in certain areas of the United States (Jamnback 1973, Cupp 1977, Stoltz 1981). A 24C Registration from the State for both pesticides was applied for and received during 1979, but the Abate label was withdrawn in October, 1979 because the Environmental Protection Agency didn't feel they had sufficient data on residues of Abate on edible plants. Dosages used by Wallace et al. (1973) in Canada were 0.1 ppm metered into streams for 15 minutes. Very limited testing of *Bacillus thuringiensis israelensis* has taken place on an isolated fast moving mountain stream. Data are not now sufficient to say how valuable it may be to the program but it is under serious consideration. Time and budget constraints have limited work on this to date.

Concern over pesticide damage to our stream environment is very real. Many papers have been written warning of the effects of Abate, Methoxychlor and other similar pesticides on non-target organisms in aquatic ecosystems that also support black fly populations. As noted above, sampling and identifying of many of these organisms have already been done and other more detailed and quantitative studies are planned for the future. The need to protect the aquatic environment while controlling black flies is a challenging problem. Until more detailed information is available, control procedures will continue with extreme caution and constant monitoring. In depth studies, such as the Athabasca reports, indicate the degree of difficulty in trying to interpret the living, moving dynamics of a large stream or river with quite differing interpretations being arrived at from the same data (Haufe 1980). As the budget will allow and with knowledge that every necessary precaution judged necessary by many authors will be observed, the District will continue with the control program and will continue to collect and evaluate data that will add to existing knowledge.

SUMMARY AND CONCLUSIONS.—Beginning in 1967, the black fly *Simulium vittatum* along with several other species has become an increasing problem in the Salt Lake Valley. Under the South Salt Lake County Mosquito Abatement Board's direction, surveys were conducted and a full scale control program was initiated in 1981. All moving water sources in the County area that produce black flies were subjected to treatment with Methoxychlor at the recommended dosage of .3 ppm/15 minute intervals. Samples have been taken of the black fly larvae in most of the streams and the Jordan River and are in the process of being identified. Square foot samples of the Jordan River bottom and two other large streams, Big and Little Cottonwood Creeks have been taken for non-target organisms and the organisms identified. A successful control program is now in operation but financial constraints and environmental considerations require a cautious expansion.

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ELECTION FOR A SPECIAL TAX RATE

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This past year three mosquito abatement districts successfully went to an election of the people to set up a special tax rate for future funding. The three districts were Colusa, Turlock, and the San Joaquin County Mosquito Abatement District. Our District, the San Joaquin County Mosquito Abatement District covers an area of approximately 1,446 square miles and has a population of over 350,000 people with a budget in 1981-82 of $1,500,000. In a district of this size and with the variety of industry, commercial, and agriculture land that we have, it made implementing an election for a special tax rate charge a very difficult and time consuming job.

The purpose of the election was to restore a major portion of money taken away through the passage of Proposition 13 in 1978.

Our Board of Trustees had a difficult time deciding whether or not to go to the election for the Special Tax rate because of the tremendous cost involved. It was the feeling of our Board of Trustees and Management that the District has the responsibility to do everything within its power to protect the residents of San Joaquin County from disease and pest mosquitoes. For that reason the Board passed a resolution to proceed and set a date for the Special Tax rate election.

Once a district decides to go the election route for funding, you must do the following:

1. Set a date for the election.
2. Choose which type of election to use.
3. The method of charge that would best suit your district.

There are three types of election procedures to choose from:

A. General Election:
   1. Least expensive election.
   2. Piggyback - share the cost with other agencies on the ballot such as: cities, counties, and state.

B. Mail Ballot Election.
   1. Ballots are sent to eligible voters by mail.
   2. Valid only in districts with 5,000 or less registered voters.
   3. Must pass by 2/3 majority of the returned ballots.

C. Special Election:
   1. Only your district is involved.
   2. Your district pays the entire cost of the election.
   3. Most expensive of the three types of elections.

There are two types of tax rate charges available:

A. Per parcel charge.
   1. A flat rate charge per parcel of land or household.

B. A charge on the square footage of land per parcel. This procedure is possible only if the county assessors office has the square footage information available.

The County has all taxable and non-taxable property listed into special "use codes". The different "use codes" are then grouped into catagories. For example; each of the below categories were assigned a tax rate charge. Our district used the following rate charges.

Maximum Charges Per Year

Residential home: 8,000 square foot lot	$1.75 per lot
Residential home on 1 acre lot	$9.56 per acre lot
Institutional - Apartments	$ 9.58 per acre
Commercial - Industrial	$ 12.77 per acre
Irrigated agriculture	$ .32 per acre
Dry farming	$ .15 per acre

There are certain election procedures you must follow:

1. A resolution must be passed by the Board of Trustees to have an election and set a date.
2. In our county the resolution setting the election date must be at least 90 days prior to the election.

3. You must set maximum rate charges to be used.
4. You must advertise in major newspapers to announce a public hearing about the election and publish your rate charges.
5. Hold the public hearing to explain the rate charges, and explain the purpose of holding the election.
6. A letter must be written to be printed in the election ballot explaining the purpose of the election and showing your rate charges.

Because of the vagueness in guidelines available to set up an election of this nature, I strongly recommend you work through county council or an attorney you are familiar with, or both.

Some methods we used for gaining public support were:

A. Speaking before service clubs.
B. Appearing before the Board of Supervisors, and City Councils requesting their endorsements, and support.
C. Working with the Farm Bureau and agriculture industry.
D. We also spoke before and received endorsements from; the Taxpayers Association, medical groups, League of Women Voters, political parties and local legislative representatives.

Public funds may not be used to advertise for the election such as; newspaper articles, brochures, paid radio or television time, etc. But you can use public funds for the actual cost of the election.

Committees can be formed for the purpose of raising money to be used for advertising purposes. Paid newspaper ads and printed brochures played an important part in our campaign.

Because of the time allowed, I have only touched on some of the procedures that are necessary when putting an election together. If any district is considering going to an election and would like further information feel free to contact me, and I would be glad to work with you.

SURVEILLANCE FOR ARTHROPOD-BORNE VIRAL ACTIVITY AND DISEASE IN CALIFORNIA DURING 1981

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This is the twelfth annual report on arthropod-borne virus surveillance in California since 1969 when testing for viruses in mosquito vectors was renewed as a surveillance activity. The report reflects the close cooperation between the Vector Biology and Control Section and the Viral and Rickettsial Disease Laboratory Section (VRDLS) of the California Department of Health Services, and the University of California, Berkeley, School of Public Health's Arthropod-Borne Virus Research Unit (UCBSPH, AVRU), along with the important contributions of the local Mosquito Abatement Districts (MADs), County Health Departments, the California Department of Food and Agriculture, private physicians and veterinarians, and others.

During 1981, there were 306 patients suspected of having encephalitis who had serum samples tested for western equine encephalomyelitis (WEE) and St. Louis encephalitis (SLE) antibodies by the State's VRDLS and the six county public health virus laboratories which assist in this phase of the surveillance effort (Table 1). As usual, a portion of these sera will also be tested by the UCBSPH, AVRU for antibodies against a variety of other arboviruses besides WEE and SLE in order to see if they might also be causes of human disease.

In addition to the serologic survey, 10 human brain samples were tested in suckling mice, but no arboviruses were isolated. No human cases of WEE or SLE were confirmed during the year. This was the second year in a row in which no human cases were proven to occur.

Serum samples from 26 clinically suspect equine cases were tested for WEE antibodies, nearly equalling the record low number during the 1980 surveillance year. These specimens were from 17 counties and one other state (Table 2). One case of WEE was confirmed: a 5 year old unvaccinated mare from Sacramento County with onset October 5; blood samples taken October 5 and October 19 showing rising WEE complement-fixing antibody titers of 1:64 to 1:512, indirect immunofluorescence titers of 1:512 and 1:512, and hemagglutination-inhibition antibody titers of 1:160 and 1:160. There were also 13 equine brain samples from 11 counties tested in suckling mice, with negative results for WEE virus. One horse brain yielded an isolate of Main Drain virus, a virus previously known to infect equines (from serologic studies) but not previously isolated from any equine. This case will be reported in detail separately.

Extensive surveillance of mosquitoes for the presence of virus was again possible this year, due to the help of the UCBSPH, AVRU in assigning Ms. Eve Tolmach to assist in the VRDLS testing program. In total, 104, 439 mosquitoes (in 2,205 pools) were tested from 23 California counties and one Arizona County (Table 3). As usual, sampling was directed most intensively to the two most important vector species (*Culex tarsalis* and *Culex pipiens* complex which represented 92.3 percent of the pools. There were 224 viral isolates made (Tables 4 and 5): 48 WEE, 2 SLE, 100 Hart Park, 65 Turlock, 8 California encephalitis group, and one as yet unidentified agent.

Sentinel chicken flocks were located at 22 sites in the state (Table 6). Seroconversions for WEE antibody were detected by the indirect fluorescent antibody method in 11 of the flocks by season's end, and for SLE antibody in 3 flocks.

Timely surveillance reports by telephone were made to the appropriate MADs or other agencies, and a weekly summary of results was again widely distributed to participants in the program. (25 issues from May 8 to October 23, 1981).

The prospects for 1982 are that high rainfall and snowfall during the 1981/82 winter will result in high run-off and the potential for excessive mosquito breeding. Despite continued budgetary restrictions, extensive surveillance and control efforts will again be needed to anticipate and prevent occurrence of epidemic arbovirus encephalitis activity in California.

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Table 1. Humans tested serologically for mosquito-borne arbovirus disease by the Viral and Rickettsial Disease Laboratory Section, California State Department of Health Services and by County Health Department laboratories, by county and month of illness onset, California, 1981.

County	Totals	Jan	Feb	March	April	May	June	July	Aug	Sept	Oct	Nov	Dec	Unknown
Totals	306	3	0	2	7	23	43	58	57	48	26	18	3	18
Alameda	1	0	0	C	0	0	1	0	0	0	0	0	0	0
Berkeley	3	0	0	0	0	1	0	0	1	1	0	0	0	0
Butte	17	1	0	1	3	3	0	0	4	4	0	1	0	0
Colusa	2	0	0	0	0	1	0	0	0	0	1	0	0	0
Contra Costa	1	0	0	0	1	0	0	0	0	0	0	0	0	0
El Dorado	10	0	0	0	0	1	0	2	1	2	2	0	0	2
Fresno*	71	0	0	0	0	0	12	30	18	6	4	1	0	0
Humboldt	4	0	0	0	0	2	0	0	0	0	1	0	0	1
Imperial	3	0	0	0	0	0	0	0	0	0	0	1	0	2
Inyo	2	0	0	0	0	0	0	0	2	0	0	0	0	0
Kings	3	0	0	0	0	0	0	0	1	2	0	0	0	0
Lassen	1	0	0	0	0	1	0	0	0	0	0	0	0	0
Los Angeles*	17	0	0	0	0	4	3	2	3	4	1	0	0	0
Mendocino	4	0	0	0	0	0	0	0	0	0	0	2	2	0
Merced	6	0	0	0	0	2	0	1	0	0	1	0	0	2
Napa	4	0	0	0	0	0	2	0	1	0	0	0	0	1
Placer	1	0	0	0	0	0	1	0	0	0	0	0	0	0
Plumas	1	0	0	0	0	0	0	0	1	0	0	0	0	0
Riverside	2	0	0	0	0	0	0	0	1	0	0	0	0	1
Sacramento*	19	0	0	1	0	0	3	1	3	4	5	1	0	1
San Bernardino*	27	0	0	0	0	0	2	1	5	8	4	6	0	1
San Diego*	48	1	0	0	0	2	9	11	8	8	4	3	0	2
San Francisco	3	1	0	0	0	1	0	1	0	0	0	0	0	0
San Joaquin	11	0	0	0	1	1	3	3	0	3	0	0	0	0
San Luis Obispo	5	0	0	0	1	1	0	1	1	0	1	0	0	0
San Mateo	1	0	0	0	0	0	0	0	0	0	0	0	0	1
Santa Barbara	7	0	0	0	0	0	0	0	2	5	0	0	0	0
Santa Clara	1	0	0	0	0	0	1	0	0	0	0	0	0	0
Santa Cruz	9	0	0	0	0	0	3	2	3	0	0	0	0	1
Shasta	4	0	0	0	0	3	0	0	0	0	0	0	1	0
Sonoma	1	0	0	0	0	0	1	0	0	0	0	0	0	0
Sutter	2	0	0	0	0	0	0	1	0	0	0	0	0	1
Tehama	4	0	0	0	0	0	1	0	0	1	0	1	0	1
Tulare	1	0	0	0	0	0	0	0	0	0	0	0	0	1
Yolo	8	0	0	0	1	0	0	2	1	0	2	2	0	0
Out of State	2	0	0	0	0	0	1	0	1	0	0	0	0	0

*Most sera tested by County Health Department Laboratory.

Table 2. Horses tested serologically for Western equine encephalomyelitis by the Viral and Rickettsial Disease Laboratory Section, California Department of Health Services, by County and month of illness onset, California, 1981.

County	Totals	Jan	Feb	March	April	May	June	July	Aug	Sept	Oct	Nov	Dec
Totals	26	0	0	0	1	2	3	1	5	5	7	2	0
Contra Costa	1	0	0	0	0	0	0	0	0	0	1	0	0
El Dorado	1	0	0	0	0	0	0	0	0	0	1	0	0
Fresno	1	0	0	0	0	0	0	0	0	0	1	0	0
Imperial	1	0	0	0	0	0	0	0	1	0	0	0	0
Kern	5	0	0	0	0	1	1	0	1	0	1	1	0
Modoc	1	0	0	0	0	0	1	0	0	0	0	0	0
Nevada	1	0	0	0	0	0	0	0	0	1	0	0	0
Orange	1	0	0	0	0	0	0	0	1	0	0	0	0
Placer	1	0	0	0	0	0	0	0	0	1	0	0	0
Plumas	2	0	0	0	0	0	0	0	0	2	0	0	0
Sacramento	2	0	0	0	0	0	0	1	0	0	1*	0	0
San Bernardino	2	0	0	0	0	0	0	0	1	0	1	0	0
Shasta	1	0	0	0	0	1	0	0	0	0	0	0	0
Siskiyou	1	0	0	0	0	0	0	0	0	1	0	0	0
Solano	1	0	0	0	0	0	0	0	0	0	0	1	0
Stanislaus	2	0	0	0	1	0	0	0	1	0	0	0	0
Ventura	1	0	0	0	0	0	1	0	0	0	0	0	0
Out of State	1	0	0	0	0	0	0	0	0	0	1**	0	0

*Positive for WEE

**Nevada

In addition, 13 equine brain samples were tested for virus by suckling mouse inoculation, with negative results for WEE Virus: 2 each from Orange County and San Joaquin Co., and 1 each from Butte, Kern, Monterey, Nevada, Sacramento, San Bernardino, Shasta, Siskiyou, and Stanislaus Counties. One of the equine brains from San Joaquin County yielded an isolate of Main Drain Virus.

Table 3. Number of mosquitoes (pools) tested, by county and species, by the Viral and Rickettsial Disease Laboratory Section, California Department of Health Services, 1981

County	TOTAL	Culex tarsalis	Culex pipiens	Aedes melanimon	Other
Butte	8,561 (177)	8,256 (168)	99 (3)	189 (5)	17 (1) Aedes vexans
Fresno	390 (10)	390 (10)			
Imperial	5,764 (128)	4,199 (93)	1,565 (35)		
Inyo	180 (7)	180 (7)			
Kern	34,707 (703)	28,111 (567)	158 (4)	6,422 (131)	16 (1) Aedes nigromaculis
Los Angeles	115 (3)	115 (3)			
Marin	508 (16)	467 (15)	41 (1)		
Merced	1,719 (38)	1,509 (33)	110 (3)	100 (2)	
Orange	970 (20)	320 (7)	650 (13)		
Placer	3,399 (70)	3,399 (70)			
Riverside	6,988 (144)	6,718 (137)	20 (2)		250 (5) Psorophora columbiae
Sacramento	1,481 (34)	1,481 (34)			
San Bernardino	2,093 (48)	2,093 (48)			
San Diego	230 (5)	230 (5)			
San Joaquin	2,068 (42)	2,068 (42)			
Shasta	1,852 (39)	1,838 (38)	14 (1)		
Sonoma	646 (15)	285 (7)	11 (1)		350 (7) Culex peus
Stanislaus	3,653 (94)	3,153 (78)	127 (4)	292 (8)	81 (4) Culex peus
Sutter/Yuba	13,024 (265)	13,024 (265)			
Tehama	124 (3)	124 (3)			
Tulare	14,138 (302)	13,838 (296)		300 (6)	
Yolo	1,609 (34)	1,609 (34)			
Mohave, AZ	220 (8)	220 (8)			
TOTAL	104,439 (2,205)	93,627 (1,968)	2,795 (67)	7,303 (152)	431 (11) Culex peus 17 (1) Aedes vexans 16 (1) Aedes nigromaculis 250 (5) Ps. columbiae

Table 4. Summary of viral isolates from mosquitoes during 1981 by the Viral and Rickettsial Disease Laboratory Section, Department of Health Services

		Viruses isolated (Minimum infection rate /1000)[1]					
County	Total	WEE	SLE	Hart Park	Turlock	California encephalitis group	Unidentified
Butte	13	2(0.2)		7(0.8)	4(0.5)		
Imperial	2	1(0.2)			1(0.2)		
Kern	88	18(0.6)		46(1.6)	15(0.5)	8(1.2)	1
Merced	2			1(0.7)	1(0.7)		
Placer	6	1(0.3)		3(0.9)	2(0.6)		
Riverside	18	17(2.5)		1(0.1)			
Sacramento	1	1(0.7)					
San Bernardino	8		2(1.0)	1(0.5)	5(2.4)		
San Joaquin	1				1(0.5)		
Stanislaus	4			2(0.6)	2(0.6)		
Sutter/Yuba	22	4(0.3)		2(0.2)	16(1.2)		
Tulare	57	4(0.3)		35(2.5)	18(1.3)		
Yolo	1			1(0.6)			
Mohave, AZ	1			1(4.5)			
Total	224	48(0.5)	2(1.0)	100(1.1)	65(0.7)	8(1.2)	1

1 - All CEV from Ae. melanimon;

All others from Cx. tarsalis

Table 5. Viral isolates from mosquito pools tested during 1981 by the Viral and Rickettsial Disease Laboratory Section, California Department of Health Services.

Identifying Number	County	Place	Date	Species	Number in Pool	Virus Isolated
E-020064	Butte	Honcut	06-30	Cx. tarsalis	50	Hart Park
E-020071	Butte	Honcut	06-30	Cx. tarsalis	50	Hart Park
E-020060	Butte	Gridley	06-23	Cx. tarsalis	50	Hart Park
E-020075	Butte	Honcut	07-07	Cx. tarsalis	50	Hart Park
E-020079	Butte	Honcut	07-07	Cx. tarsalis	50	Hart Park
E-020084	Butte	Gridley	07-07	Cx. tarsalis	50	Hart Park
E-020102	Butte	Gridley	07-14	Cx. tarsalis	50	Turlock
E-020108	Butte	Nord	07-21	Cx. tarsalis	50	WEE
E-020121	Butte	Honcut	07-28	Cx. tarsalis	50	Turlock
E-020124	Butte	Gridley	07-28	Cx. tarsalis	50	Turlock
E-020131	Butte	Gridley	07-28	Cx. tarsalis	50	Turlock
E-020171	Butte	Honcut	08-18	Cx. tarsalis	50	Hart Park
E-020193	Butte	Nord	09-01	Cx. tarsalis	50	WEE
E-000017	Imperial	Bard	06-09	Cx. tarsalis	48	Turlock
E-000039	Imperial	Calipatria	06-08	Cx. tarsalis	50	WEE
E-018645	Kern	Buttonwillow	06-02	Cx. tarsalis	50	Hart Park
E-018678	Kern	Buttonwillow	06-09	Cx. tarsalis	50	Hart Park
E-018713	Kern	Buttonwillow	06-23	Cx. tarsalis	45	Hart Park
E-018717	Kern	Delano	06-23	Ae. melanimon	50	CEV
E-006367	Kern	City Sewer Farm	06-23	Cx. tarsalis	50	Hart Park
E-006368	Kern	City Sewer Farm	06-23	Cx. tarsalis	50	Hart Park
E-006372	Kern	City Sewer Farm	06-23	Cx. tarsalis	50	Hart Park
E-018731	Kern	Bakersfield	06-24	Cx. tarsalis	50	Hart Park
E-018751	Kern	Bakersfield	07-01/02	Cx. tarsalis	29	Hart Park
E-006382	Kern	City Sewer Farm	07-07	Cx. tarsalis	50	Hart Park
E-006388	Kern	City Sewer Farm	07-07	Cx. tarsalis	50	Hart Park
E-006391	Kern	City Sewer Farm	07-07	Cx. tarsalis	50	Hart Park
E-006392	Kern	City Sewer Farm	07-07	Cx. tarsalis	50	Hart Park
E-006395	Kern	City Sewer Farm	07-07	Cx. tarsalis	50	Turlock
E-006397	Kern	City Sewer Farm	07-14	Cx. tarsalis	50	Hart Park
E-006402	Kern	Jessup Dairy	07-14	Cx. tarsalis	50	Hart Park
E-006403	Kern	Jessup Dairy	07-14	Cx. tarsalis	50	Hart Park
E-006408	Kern	Fish Ranch	07-14	Cx. tarsalis	50	Hart Park
E-006409	Kern	Fish Ranch	07-14	Cx. tarsalis	50	Hart Park
E-006410	Kern	Fish Ranch	07-14	Cx. tarsalis	50	Hart Park
E-018785	Kern	Buttonwillow	07-14	Cx. tarsalis	50	Hart Park
E-018786	Kern	Buttonwillow	07-14	Cx. tarsalis	50	Hart Park
E-006419	Kern	Twin Farms	07-21	Ae. melanimon	50	CEV
E-018799	Kern	Delano	07-21/22	Cx. tarsalis	39	Turlock
E-018802	Kern	Buttonwillow	07-21/22	Cx. tarsalis	50	Hart Park
E-018808	Kern	Bakersfield	07-21/22	Cx. tarsalis	50	Hart Park
E-018809	Kern	Bakersfield	07-21/22	Cx. tarsalis	50	Hart Park
E-006430	Kern	Costerison's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006431	Kern	Costerison's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006432	Kern	Costerison's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006435	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Turlock
E-006441	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006442	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006445	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006448	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Hart Park
E-006450	Kern	Garone's Farm	07-28	Cx. tarsalis	50	Hart Park
E-021107	Kern	J.T.Sanders Duck Club	08-04	Cx. tarsalis	50	Hart Park
E-018840	Kern	Buttonwillow	08-04/05	Cx. tarsalis	50	Hart Park
E-021114	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Turlock
E-021116	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park

Table 5. (continued)

Identifying Number	County	Place	Date	Species	Number in Pool	Virus Isolated
E-021117	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park
E-021120	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park
E-021122	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park
E-021128	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Turlock
E-021132	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park
E-021133	Kern	Garone's Farm	08-11	Cx. tarsalis	50	Hart Park
E-018863	Kern	Buttonwillow	08-11/12	Cx. tarsalis	50	Turlock
E-018868	Kern	Delano	08-11/12	Cx. tarsalis	50	Hart Park
E-018897	Kern	Buttonwillow	08-18/19	Cx. tarsalis	50	Turlock
E-018898	Kern	Buttonwillow	08-18/19	Cx. tarsalis	50	Hart Park
E-18900	Kern	Buttonwillow	08-18/19	Ae. melanimon	50	CEV
E-021140	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021141	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021142	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021143	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021144	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021146	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	Hart Park
E-021147	Kern	Tracy Ranch	08-18	Cx. tarsalis	50	WEE
E-021154	Kern	Eureka Duck Club	08-18	Cx. tarsalis	50	Hart Park
E-021188	Kern	Bakersfield	08-18	Cx. tarsalis	50	WEE
E-021189	Kern	Bakersfield	08-18	Cx. tarsalis	50	Hart Park
E-018910	Kern	Delano	08-25/26	Cx. tarsalis	50	Turlock
E-018924	Kern	Buttonwillow	08-25/26	Ae. melanimon	50	CEV
E-018925	Kern	Buttonwillow	08-25/26	Ae. melanimon	50	CEV
E-018945	Kern	Buttonwillow	09-01/02	Cx. tarsalis	50	Hart Park
E-021216	Kern	Bakersfield	09-09	Cx. tarsalis	50	Turlock
E-021218	Kern	Bakersfield	09-09	Cx. tarsalis	50	WEE
E-021220	Kern	Bakersfield	09-09	Cx. tarsalis	50	WEE
E-021231	Kern	Arvin	09-09	Cx. tarsalis	50	WEE
E-014147	Kern	Delano	09-14	Cx. tarsalis	50	Turlock
E-014149	Kern	Delano	09-14	Cx. tarsalis	50	WEE
*E-014153	Kern	Delano	09-14	Cx. tarsalis	50	Unidentified agent
E-014162	Kern	Delano	09-14	Cx. tarsalis	50	Turlock
E-018966	Kern	Bakersfield	09-09/10	Cx. tarsalis	50	Turlock
E-018973	Kern	Buttonwillow	09-09/10	Cx. tarsalis	50	WEE
E-018978	Kern	Buttonwillow	09-09/10	Cx. tarsalis	50	WEE
E-018979	Kern	Buttonwillow	09-09/10	Cx. tarsalis	50	WEE
E-018989	Kern	Buttonwillow	09-09/10	Ae. melanimon	50	CEV
E-018993	Kern	Delano	09-09/10	Cx. tarsalis	50	WEE
E-021241	Kern	Wasco	09-15	Cx. tarsalis	50	WEE
E-021258	Kern	Wasco	09-15	Cx. tarsalis	50	WEE
E-019032	Kern	Buttonwillow	09-15/16	Ae. melanimon	50	CEV
E-019040	Kern	Buttonwillow	09-22/23	Cx. tarsalis	50	Turlock
*E-019041	Kern	Buttonwillow	09-22/23	Cx. tarsalis	50	Turlock & Hart Park
E-019044	Kern	Buttonwillow	09-22/23	Cx. tarsalis	50	WEE
E-019046	Kern	Buttonwillow	09-22/23	Ae. melanimon	50	CEV
E-019049	Kern	Buttonwillow	09-22/23	Cx. tarsalis	50	Turlock
E-016973	Merced	Merced	07-14	Cx. tarsalis	50	Turlock
E-016974	Merced	Merced	07-14	Cx. tarsalis	50	Hart Park
E-010721	Placer	Roseville	07-06	Cx. tarsalis	50	Hart Park
*E-010738	Placer	Sheridan	07-06	Cx. tarsalis	50	Hart Park & Turlock
E-010741	Placer	Sheridan	07-06	Cx. tarsalis	50	Hart Park
E-010781	Placer	Roseville	07-16	Cx. tarsalis	50	Turlock
E-010789	Placer	Rocklin	07-20	Cx. tarsalis	50	WEE
E-002847	Riverside	Mecca	05-19	Cx. tarsalis	50	WEE

Table 5. (continued)

Identifying Number	County	Place	Date	Species	Number in Pool	Virus Isolated
E-002852	Riverside	Mecca	05-19	Cx. tarsalis	50	Hart Park
E-002854	Riverside	Mecca	05-19	Cx. tarsalis	50	WEE
E-002864	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002865	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002867	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002868	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002869	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002870	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002872	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002874	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002875	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002877	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002878	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002880	Riverside	Mecca	06-16	Cx. tarsalis	50	WEE
E-002896	Riverside	Mecca	07-22	Cx. tarsalis	50	WEE
E-002898	Riverside	Mecca	07-22	Cx. tarsalis	50	WEE
E-003200	Riverside	Mecca	07-22	Cx. tarsalis	50	WEE
E-010818	Sacramento	Sacramento	07-09	Cx. tarsalis	50	WEE
E-002960	San Bernardino	Parker	06-10	Cx. tarsalis	50	Turlock
E-002962	San Bernardino	Needles	07-22	Cx. tarsalis	42	Hart Park
E-002964	San Bernardino	Needles	07-22	Cx. tarsalis	50	Turlock
E-002969	San Bernardino	Needles	07-22	Cx. tarsalis	50	Turlock
E-002970	San Bernardino	Needles	07-22	Cx. tarsalis	50	Turlock
E-002971	San Bernardino	Needles	07-22	Cx. tarsalis	10	Turlock
E-002976	San Bernardino	Needles	08-20	Cx. tarsalis	50	SLE
E-002977	San Bernardino	Needles	08-20	Cx. tarsalis	50	SLE
E-010771	San Joaquin	Escalon	07-09	Cx. tarsalis	50	Turlock
E-010040	Stanislaus	Denair	06-30	Cx. tarsalis	29	Hart Park
E-010755	Stanislaus	Valley Home	07-09	Cx. tarsalis	50	Hart Park
E-010053	Stanislaus	Newman	08-04/05	Cx. tarsalis	50	Turlock
E-010067	Stanislaus	Newman	08-04	Cx. tarsalis	16	Turlock
E-020253	Sutter/Yuba	Wheatland	06-22	Cx. tarsalis	50	Turlock
E-020268	Sutter/Yuba	Live Oak	06-23	Cx. tarsalis	50	Turlock
E-020274	Sutter/Yuba	Yuba City	06-23	Cx. tarsalis	50	Turlock
E-020275	Sutter/Yuba	Yuba City	06-23	Cx. tarsalis	50	Turlock
E-020287	Sutter/Yuba	Linda	06-29	Cx. tarsalis	50	Turlock
E-020290	Sutter/Yuba	Marysville	06-29	Cx. tarsalis	50	Hart Park
E-020299	Sutter/Yuba	Live Oak	06-30	Cx. tarsalis	50	Turlock
E-020304	Sutter/Yuba	East Nicolaus	06-30	Cx. tarsalis	50	Hart Park
E-020305	Sutter/Yuba	Yuba City	06-30	Cx. tarsalis	50	Turlock
E-020330	Sutter/Yuba	Yuba City	07-06	Cx. tarsalis	50	Turlock
E-020332	Sutter/Yuba	Marysville	07-06	Cx. tarsalis	50	Turlock
E-020337	Sutter/Yuba	Yuba City	07-13	Cx. tarsalis	50	Turlock
E-020349	Sutter/Yuba	Live Oak	07-20	Cx. tarsalis	50	Turlock
E-020364	Sutter/Yuba	Marysville	07-21	Cx. tarsalis	50	Turlock
E-020368	Sutter/Yuba	Marysville	07-21	Cx. tarsalis	50	Turlock
E-020394	Sutter/Yuba	Live Oak	07-28	Cx. tarsalis	50	Turlock
E-020431	Sutter/Yuba	Yuba City	08-10	Cx. tarsalis	50	WEE
E-020442	Sutter/Yuba	Marysville	08-10	Cx. tarsalis	50	Turlock
E-020476	Sutter/Yuba	Yuba City	08-25	Cx. tarsalis	50	Turlock
E-020489	Sutter/Yuba	Yuba City	08-31	Cx. tarsalis	50	WEE
E-018561	Sutter/Yuba	Meridan	09-01	Cx. tarsalis	50	WEE
E-018572	Sutter/Yuba	Meridan	09-08	Cx. tarsalis	50	WEE
E-018135	Tulare	Teviston	07-06	Cx. tarsalis	50	Hart Park
E-018153	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018138	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park

Table 5. (continued)

Identifying Number	County	Place	Date	Species	Number in Pool	Virus Isolated
E-018139	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018140	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018142	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018145	Tulare	Teviston	07-21	Cx. tarsalis	50	Turlock
E-018146	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018147	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018148	Tulare	Teviston	07-21	Cx. tarsalis	50	Hart Park
E-018159	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018160	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018162	Tulare	Teviston	08-03	Cx. tarsalis	50	Turlock
E-018163	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018164	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018175	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018180	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018183	Tulare	Teviston	08-03	Cx. tarsalis	50	Turlock
E-018185	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018186	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-018187	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-021001	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-021003	Tulare	Teviston	08-03	Cx. tarsalis	50	Turlock
E-021005	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-021006	Tulare	Teviston	08-03	Cx. tarsalis	50	Turlock
E-021007	Tulare	Teviston	08-03	Cx. tarsalis	50	Hart Park
E-021021	Tulare	Earlimart	08-10	Cx. tarsalis	50	Hart Park
E-021025	Tulare	Earlimart	08-10	Cx. tarsalis	50	Turlock
E-021028	Tulare	Earlimart	08-10	Cx. tarsalis	50	Hart Park
E-021029	Tulare	Earlimart	08-10	Cx. tarsalis	18	Turlock
E-021033	Tulare	Teviston	08-10	Cx. tarsalis	50	Turlock
E-021034	Tulare	Teviston	08-10	Cx. tarsalis	50	Hart Park
E-021039	Tulare	Earlimart	08-10	Cx. tarsalis	50	Hart Park
E-021048	Tulare	Earlimart	08-10	Cx. tarsalis	50	Hart Park
E-021049	Tulare	Earlimart	08-10	Cx. tarsalis	50	Hart Park
E-021051	Tulare	Teviston	08-10	Cx. tarsalis	50	Turlock
E-021052	Tulare	Teviston	08-10	Cx. tarsalis	50	Turlock
E-021053	Tulare	Teviston	08-10	Cx. tarsalis	50	Hart Park
E-021060	Tulare	Pond	08-17	Cx. tarsalis	50	Hart Park
E-006160	Tulare	Pixley	08-18	Cx. tarsalis	50	Turlock
E-006161	Tulare	Pixley	08-18	Cx. tarsalis	55	Hart Park
E-006162	Tulare	Pixley	08-18	Cx. tarsalis	60	Turlock
E-006163	Tulare	Pixley	08-18	Cx. tarsalis	60	Hart Park
E-021069	Tulare	Teviston	08-24	Cx. tarsalis	50	Turlock
E-021070	Tulare	Teviston	08-24	Cx. tarsalis	50	Hart Park
E-021071	Tulare	Teviston	08-24	Cx. tarsalis	50	Hart Park
E-021088	Tulare	Teviston	08-24	Cx. tarsalis	50	Turlock
E-021090	Tulare	Teviston	08-24	Cx. tarsalis	50	Turlock
E-021099	Tulare	Earlimart	08-24	Cx. tarsalis	50	Hart Park
E-021346	Tulare	Earlimart	08-24	Cx. tarsalis	50	Turlock
E-021349	Tulare	Earlimart	08-24	Cx. tarsalis	50	Hart Park
E-014124	Tulare	Teviston	09-08	Cx. tarsalis	50	WEE
E-014131	Tulare	Delano	09-08	Cx. tarsalis	50	WEE
E-014134	Tulare	Delano	09-08	Cx. tarsalis	50	Turlock
E-014139	Tulare	Teviston	09-14	Cx. tarsalis	50	WEE
E-014141	Tulare	Teviston	09-14	Cx. tarsalis	50	Turlock
E-014142	Tulare	Teviston	09-14	Cx. tarsalis	50	WEE
E-010825	Yolo	Woodland	07-16	Cx. tarsalis	50	Hart Park
E-002979	Mohave, AZ	Needles	08-20	Cx. tarsalis	28	Hart Park

Table 6. Serological conversions to WEE and SLE viruses in sentinel chickens, California, 1981.

County	Location	Number (percent)	positive		Cx. tarsalis per trap night	
		WEE	SLE	1st pos	Season	Highest (date)
Shasta	MAD office	0	0	-	3.3	10.3 (8-11)
Tehama	MAD office	2(10)	0	8-3	11.0	45.1 (9-11)
Butte	Chico	1(4)	0	8-31	0.3	2.3 (9-8)
Butte	Grey Lodge	1(5)	0	9-28	48.4	164.6 (7-14)
Colusa	MAD office	2(9)	0	9-1	12.8	118.9 (7-23)
Yuba	Marysville	3(13)	0	8-26	7.1	16.3 (9-17)
Sutter	Dean's	0	0	-	41.8	192.0 (7-8)
Sacramento	Elk Grove	0	0	-	3.7*	6.4 (8-12)
San Joaquin	Escalon	0	0	-	5.2	27.4 (7-8)
Merced	Stevinson	0	0	-	5.9	18.3 (9-17)
Fresno	Mendota Refuge	0	0	-	25.6	140.2 (9-30)
Tulare	Rocky Hill	0	0	-	0.6	1.4 (8-20)
Kern	Teviston	2(11)	0	9-28	0.5	3.0 (9-17)
Kern	Oildale	0	0	-	0.1	2.5 (4-15)
Kern	Wasco	0	0	-	0.4	3.0 (9-10)
Kern	Buttonwillow	8(33)	0	9-4	2.8	11.0 (9-2)
Kern	F.C.Tracy	10(48)	0	9-4	4.0	13.5 (8-5)
Kern	John Dale	4(17)	0	10-7	17.1	72.0 (9-2)
San Bernardino	Needles	0	0	-	20.3	138.3 (5-25)
Riverside	Mecca	6(30)	1(5)	7-3 WEE 8-5 SLE	6.0	22.7 (4-29)
Riverside	Corona	0	2(12)	10-5	0.2	1.1 (6-25)
Imperial	Corda Ranch	4(22)	5(28)	8-5 both	1.5	12.6 (7-21)

* = Data for August only

VARIATION IN THE VECTOR COMPETENCE OF *CULEX PIPIENS* COMPLEX TO ST. LOUIS ENCEPHALITIS VIRUS

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ABSTRACT

In the Central and Eastern United States, *Culex pipiens* L. and *Cx. quinquefasciatus* Say are important vectors of St. Louis encephalitis virus (SLE). Both species, however, are apparently not important vectors of SLE in the Western United States. Differences in the vector competence between eastern and central, and western populations could be attributed to, among other factors, differences in the infectibility and/or transmissibility of geographic strains of SLE virus by the indigenous mosquito populations. Present studies evaluated the vector competence (paired tests) of *Cx. pipiens* populations from California (Sacramento Valley) and Maryland, and *Cx. quinquefasciatus* populations from California (San Joaquin Valley) and Texas. Each population (F_1 progeny) was infected orally by feeding on viremic chicks that were inoculated with a representative SLE viral strain from each of the four geographical locations. Mosquitoes were tested for their ability to become infected and transmit the virus by bite after an extrinsic incubation period of 13 to 22 days at 25 ± 1°C. Results indicated that *Cx. pipiens* from Maryland were slightly more efficient transmitters of virus than were *Cx. pipiens* from the Sacramento Valley. In contrast, *Cx. quinquefasciatus* from the San Joaquin Valley were more efficient vectors compared to *Cx. quinquefasciatus* from Texas. As reported elsewhere, *Cx. pipiens* is overall a better vector of SLE than *Cx. quinquefasciatus*. Though differences were evident in the transmissibility of different viral strains evaluated, there was no consistency among the populations tested to transmit with greater efficiency either eastern and central, or western viral strains. Thus, our results provide no conclusive evidence as to why *Cx. pipiens* or *Cx. quinquefasciatus* populations in California are less important vectors of SLE than their counterparts in the Central and Eastern United States.

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CANINE HEARTWORM IN THE SAN FRANCISCO BAY AREA: EPIZOOTIOLOGY AND CONTROL

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ABSTRACT

Dirofilaria immitis Leidy, the causative agent of canine heartworm disease, is a parasitic mosquito-borne filarial nematode transmitted by several species of mosquitoes throughout the world. This serious canine disease of the Atlantic and Gulf Coast States, is now common in the Great Lakes and New England areas of the United States. In California, investigations of canine heartworm have been few and limited in scope. San Mateo County, California, experienced an epizootic level of infection in 1977 when more than 50 cases of heartworm were diagnosed by one animal clinic which prior to 1977 had identified less than 6 cases annually. It is noteworthy that *D. immitis* infections occurred predominantly in dogs native to California, not dogs introduced from enzootic areas outside California.

The epizootiology of canine heartworm was primarily investigated in three San Francisco Bay Area Counties (San Mateo, Santa Clara and Santa Cruz) during 1979, 1980 and 1981. Investigations focused on the following: role of the domestic dog; role of the wild coyote; identification of potential vector mosquitoes; and development of operational guidelines for control within enzootic areas. Three dog heartworm clinics were conducted, revealing an incidence of *D. immitis* in domestic dogs in the study area of 39 percent. This represented 63 positive cases of 160 dogs examined. The influence of age, weight, sex, neutered state and indoor versus outdoor dwelling habits of dogs were analyzed. Analysis of age factor and infection rate by regressing analysis indicated a significant positive correlation, the proportion of parasitized dogs did in fact accelerate with age. No relationship existed between the other factors and the incidence of canine heartworm. The wild coyote *Canis latrans*, a reservoir animal in the San Francisco Bay Area, exhibited an infection rate less than one-third that found in domestic dogs from the same area. Six mosquito species were considered potential vectors. Based on high adult densities and high susceptibility to heartworm infection, *Aedes sierrensis* is the most probable primary vector.

With improved coordination of canine heartworm research in California and verification of transmission by particular vector species, vector control agencies will be better prepared to concentrate control efforts where applicable. Cooperation among the public sector, control agencies and veterinarians is necessary to identify enzootic areas for heartworm and initiate procedures to minimize the impact of potential epizootics and reduce the incidence of parasitism among domestic dogs.

EFFICACY OF THE MOSQUITO FUNGAL PATHOGEN, *LAGENIDIUM GIGANTEUM* (OOMYCETES:LAGENIDIALES)

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The use of fungi for control of mosquito larvae in selected habitats is promising. There are obstacles to be overcome, however, and the use of fungi is not presently practical in an operational program. At North Carolina State University and the University of North Carolina (Aris Domnas) we are engaged in cooperative research (supported by NIH grant 1RO1 AI 17024) to develop the use of the fungi *Lagenidium giganteum*. Two isolates are being studied: North Carolina (NC) and Louisiana (LA). Following is a summary of recent results from this program.

CULTURE TECHNIQUES.–*In vitro* culturing of *Lagenidium giganteum* is being improved using both broth and solid (agar) media techniques. Presently maintenance of the fungi is best accomplished on a broth medium (WYGH) composed of wheat germ (3.2 g/liter), yeast extract (1.4 g/liter) and glucose (1.2 g/liter) with addition of sufficient aqueous extract of hemp seed to result in 0.25 mg soluble protein per ml of broth. The fungus may also be grown in PYG broth (composed of 1.25 g/liter peptone, 1.25 g/liter yeast extract and 3 g/liter glucose). The broth cultures are in flasks on a shaker table. Although more research is needed to maximize production, so far the greatest yield has been by growing the fungus in PYG broth for five-six days, then on agar plates (containing 2% agar prepared with aqueous hemp seed extract added in sufficient quantity to give 1.0 mg soluble protein per ml). After seven days on the hemp seed-agar plate the plate is placed in 1000 ml of distilled water for 18 hours (the time for peak production of vesicles and zoospores) at 27°C.

Time intervals on broth and on hemp seed-agar are critical. Methods are being tested to produce sporulation directly from the broth without using the plates, but so far the results are highly variable.

The agar plates may be stored for up to 10 days if kept moist and will remain active, i.e. the fungus will produce zoospores upon flooding with large volumes of water. For greatest production the agar in the plates should be thin (0.75-1.0 mm).

At present the most practical method for field introduction of the fungus cultured by *in vitro* methods is by using the agar plate material.

ENVIRONMENTAL EFFECTS.–For effective and practical use of the fungus in the field it is necessary to be able to predict the liklihood of successful infection of mosquito larvae by introduced fungus. Factors in the aquatic habitat which we are quantifying are: organic pollution, salinity, temperature, pH and pesticides.

Organic pollution. Sporulating agar-plate cultures of *Lagenidium giganteum* were introduced into outdoor, pilot-scale poultry-waste lagoons. In unpolluted water the fungus infected 27-29% of *Culex quinquefasciatus* larvae, depending upon the size of the fungal inoculum. *Lagenidium* persisted for one month in these habitats until low water temperatures inhibited fungal development.

Low to moderate levels of organic water pollution completely prevented infection of larvae by *Lagenidium* in the lagoons.

Laboratory tests of the effects of pollution levels on zoosporogenesis and larval infection by *Lagenidium* allowed development of a multiple regression formula that related levels of chemical oxygen demand (COD), total Kjeldhal nitrogen (TKN), ammonia-nitrogen (NH_3) and total phosphorous (P) to percentage infection of larvae under standardized, optimum conditions. The regression is % Infection = 113.9-.02 (mg/l COD) - 6.02 (mg/l P) - 3.41 (mg/l NH_3) - 1.92 (mg/l TKN). This formula means that the infection rate would decrease .02% for each additional mg/l COD, 6.02% for each additional mg/l P, 3.41% for each additional mg/l NH_3 and 1.92% for each added mg/l TKN, when all four parameters were considered simultaneously. Only NH_3 and P were statistically reliable predictors of *Lagenidium* performance in polluted water.

Observations of sporulating fungus cultures in polluted water revealed that organic pollution interferes with zoosporogensis and with viability of any zoospores that have been produced.

Our experiments indicate that *Lagenidium giganteum* has little or no potential as a biological control agent in habitats having even slight organic pollution. In unpolluted water, however, the fungus can cause epizootics and recycle itself as long as water temperatures are favorable for its growth.

Salinity. Laboratory experiments were conducted to determine the salinity range over which North Carolina (NC) and Louisiana (LA) isolates of *Lagenidium giganteum* (Couch) grow vegetatively and infect mosquito larvae. The mycelial growth rates of the two isolates on nutrient agar were increased with 2.5 and 5.0 parts per thousand (ppt) of NaCl added and reduced with 7.5 ppt or more NaCl. The LA and NC isolates did not grow on agar containing 20 and 30 ppt NaCl, respectively. The IC_{50} values for the inhibition of mycelial growth of the LA and NC isolates (10.9 and 12.0 ppt NaCl, respectively) were not significantly ($p < .05$) different.

The ability of the two isolates of *L. giganteum* to infect larvae of *Aedes taeniorhynchus* (Wiedemann) decreased as the salinity of the water increased. The IC$_{50}$ values for the inhibition of infection in mosquito larvae by the LA and NC isolates (0.52 and 0.55 ppt NaCl, respectively) were not significantly ($P < .05$) different. Microscopic examination of the fungus in saline and distilled waters showed that NaCl inhibited the production of zoospores. In water containing 1.5 ppt NaCl there was complete (100%) inhibition of zoosporogenesis and mosquito infection in each isolate of *L. giganteum*. Zoosporogenesis was ca 22 times more sensitive to salinity than was mycelial growth.

Temperature and pH. From field and laboratory experiments with the NC and LA isolates it is apparent that *Lagenidium* infection of mosquito larvae is optimal at temperatures of 21 to 29°C. If the temperature is much above 30°C for appreciable periods of time the fungus will not be effective against mosquito larvae. The fungus readily infected mosquito larvae within a pH range of 6.3-8.9 in both laboratory and field experiments.

Pesticides. Due to the use of certain insecticides for mosquito control, herbicides for weed control and contamination of aquatic habits by runoff from agricultural lands, the compatibility of *Lagenidium* with various pesticides in the aquatic environment may become important. The two isolates of *Lagenidium giganteum* were tested in the laboratory for their abilities to continue mycelial growth and to produce zoospores. Dilutions of the chemicals (technical grade) were incorporated into PYG agar at the recommended field rates and several log$_{10}$ concentrations higher. Petri dishes were inoculated and mycelial growth rates were determined. Percent inhibition of mycelial growth relative to control vs. log$_{10}$ dose were probit transformations.

The results of compatability tests of the NC isolate and the following pesticides gave the following LC$_{10}$ and LC$_{50}$ values (ppm), respectively: malathion, 20.9, 130.6; chlorpyrifos, 6.1, 569.9; temephos, 168.8, 4697.0; fenthion, 8.9, 120.6; toxaphene, 6.1, 90.8; DDT, 38.7, 1862.0; lindane, 0.9, 2.0; carbaryl, 13.4, 43.7; propoxur, 127.6, 231.3; atrazine, 13.3, 67.5; captan, 16.6, 64.7. Similar studies with LA isolate showed the following LC$_{10}$ and LC$_{50}$ values (ppm), respectively: malathion, 6.0, 32.0; chlorpyrifos, 76.1, 1439.7; temephos, 345.3, 1653.3; fenthion, 2.26, 54.3; toxaphene, 2.0, 37.4; DDT, 6.2, 586.7; lindane, 0.3, 1.5; carbaryl, 12.6, 39.4; propoxur, 58.5, 207.0; captan, 7.6, 44.9; atrazine, 15.2, 94.3. Other chemicals that appeared to be less toxic to the mycelial growth of each isolate of *L. giganteum* at the levels tested were the following: methoprene, IC$_{10}$ ca. 1000 ppm; permethrin, LC$_{10}$ > 180 ppm; dimilin, LC$_{10}$ > 740 ppm. Additional tests are currently in progress to study the effects of these and other pesticides on zoospore production.

SUMMARY.–*Lagenidium giganteum* is potentially a useful biological control agent for mosquito larvae under certain environmental conditions such as: very little or no organic pollution, water temperatures not above 30°C for any appreciable length of time, and little or no salinity. Inhibiting pesticides should not be present. In most mosquito breeding habitats the pH level will not be a limiting factor. The fungus can be routinely produced by *in vitro* methods and introduced into the field from agar-plate cultures. Under some circumstances the fungus will overwinter and recycle in a mosquito-breeding habitat.

Like any biological control agent, *Lagenidium giganteum* will be a useful tool (but not a panacea) to be considered in a mosquito IPM program. Further research and development is necessary before its use in operational IPM programs is practical.

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ARTIFICIAL CULTURE OF THE SEXUAL AND ASEXUAL STAGES OF *LAGENIDIUM GIGANTEUM*

James L. Kerwin and Robert K. Washino

Lagenidium giganteum, a facultative parasite of mosquito larvae, infects its host via motile zoospores which can be formed either sexually or asexually. Like many other oomycetous fungi, *L. giganteum* requires an exogenous source of sterols for the induction of either type of reproduction. It has been known for a number of years that sterols structurally related to cholesterol are necessary for asexual reproduction. Recently we have defined conditions under which the sexual or oospore stage can be induced *in vitro* using sterols.

Artificial culture of oospores of this fungus is potentially amenable to commerical production. Desirable characteristics of oospores include abrasion resistance, tolerance to temperature extremes, and the ability for storage in a dehydrated state for months or years without loss of viability. In addition, oospores can be disseminated using techniques commonly employed for insecticides.

Under appropriate conditions following a six to ten week maturation period, oospores grown in artificial culture will produce a germ tube from which 15 to 20 biflagellate zoospores are released. These zoospores are morphologically indistinguishable at the light microscope level from asexual zoospores.

Based on field observations and initial laboratory evaluations, zoospores generated via the sexual process appear to be much more virulent than asexual zoospores against both mosquito species, *Culex tarsalis* and *Anopheles freeborni*, associated with rice culture in California's Central Valley. Field evaluations in the 1982 season will further test the efficacy of the sexual stage of *L. giganteum* in mosquito control.

MASS REARING OF *ROMANOMERMIS CULICIVORAX*
(A PROGRESS REPORT)

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In May, 1980, Sutter Yuba Mosquito Abatement District, in cooperation with U.C. Davis, began a project to mass rear the mermithid nematode *Romanomermis culicivorax*. The first eight months focused on training district personnel and on establishing a mass colony of *Culex pipiens*, the host mosquito for the nematodes.

Since the project began, improvements have heen made in equipment and some time saving steps have been implemented. The rearing laboratory currently houses thirty 2 ft. x 2 ft. x 2 ft. adult mosquito cages and sixteen metal racks for holding fiberglass rearing pans. Each rack holds eight pans. Larvae are reared at 10,000 per pan.

Two additions to the project were recently made. A screened building was constructed outdoors to house the *Coturnix* quail used as blood hosts for the mosquitoes. At this time 120 quail are used per week. In our existing building, a temperature controlled storage room was constructed for nematode egg cultures.

With the exception of a few modifications, the basic rearing techniques for both the nematode and the mosquito remain the same. Mosquito larvae are fed a diet of finely milled commerical rat chow. In addition to a weekly blood meal, the adults are fed a 10% sucrose solution. Four days after a blood meal, egg rafts are collected and, by means of a grid system, the desired number of egg rafts are added to small containers and left to hatch before being added to the rearing pans.

Preparasites are introduced to first instar larvae at a 4:1 (nematode: mosquito) ratio. Larvae are fed daily. On the seventh day, infected larvae are collected and concentrated in plastic emergence containers. Post parasites are collected daily until emergence has finished.

New cultures are started by adding one litre of dechlorinated water to a plastic shoe box filled with one inch of autoclaved medium coarse sand. Post parasites are added at 5 grams per culture. Cultures are drained after three weeks and are stored on shelves until the eggs have matured.

During the summer and early winter months of 1981, cultures had not been stored in a temperature controlled area; temperatures varied widely and were influenced by outside seasonal temperatures. In August, when three week old cultures were routinely drained, active preparasites were found. This indicated that within the initial three week period, post parasites had matured, completed oviposition and some eggs had matured and hatched. In January, 12 week old cultures which should have contained mature eggs were flooded and yielded 0 - 10,000 preparasites. By placing these 12 week old cultures in a storage area at a constant temperature of 27°C., preparasitic production increased in three weeks, and further floodings at two week intervals produced even greater increases. Further tests with additional cultures are being made, and while results are still inconclusive, preliminary results indicate that by using controlled temperatures, cultures might be timed to meet the seasonal needs for field applications.

During the summer months of 1981, the district provided Dr. Robert Washino and his staff with post parasites for rice field mosquito control. In addition the district made post parasite applications at some overwintering test sites in drainage canals, cemetery ponds and golf course ponds during the fall.

We will be providing post parasites for U.C. Davis' continuing rice field studies this summer. The district's plans include tests for presence of parasites in last year's overwintering test sites, along with new sites to be tested.

This year's post parasite production has already exceeded 1981's production. Preliminary designing has begun for expanding our rearing facilities. The planned addition to the existing building will add two to three times the area to the present rearing lab.

Throughout the past year we appreciated the guidance and assistance of Dr. Robert Washino and his staff, Dr. Edward Platzer and Dr. Becky Westerdahl.

NEW PARASITIC INSECTS FOR BIOLOGICAL CONTROL OF SYNANTHROPIC FLIES[1]

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Parasitic insects contribute significantly to the natural biological reduction of synanthropic flies of the endophilous category, where abundance is wholly dependent on human activity (Legner et al. 1974). Control of this group of flies, including the common house fly and several species of *Fannia*, can often be increased by inundation with parasitic insects, the most effective being species of *Spalangia, Muscidifurax* and occasionally *Pachycrepoideus vindemiae* Rondani and *Tachinaephagus zealandicus* Ashmead (Legner 1981, Legner and Brydon 1966, Legner and Dietrick 1972, Morgan 1981, Morgan et al. 1975a, 1975b, 1976, Olton and Legner 1975, Rutz and Axtell 1979).

Although parasite inundation combined with sound filth habitat management (Legner et al. 1973) may reduce fly abundance to below the annoyance threshold, it is apparent that degrees of control may vary in different climates. The parasitic insects that have been available for inundation have until now been restricted to single strains of a few species, usually those acquired at subtropical latitudes where initial experiments were conducted to show their effectiveness.

Current investigations show that fly parasite strains and species from different geographical and climatic areas differ in their activity, a finding that is becoming increasingly apparent in other host-parasite relationships. The utilization of some of these new species and strains which are capable of greater parasitization rates in a wider variety of climates offers possibilities for substantial gains in the biological control of synanthropic flies. This reassessment involving parasites of *Musca domestica* L., *Stomoxys calcitrans* (L.), *Fannia canicularis* (L.), and *Fannia femoralis* Stein, pointed to the absence of several noteworthy species and strains which attack these pests in other parts of the world or in isolated areas of the North American Continent. Some of the new parasites possess characteristics suited to a broader climatic range such as is found in the central and western United States where excessive heat and drought often restrict the performance of resident species or those that were available through commercial sources.

For example, the parasite *Sphegigaster* sp. (FIG. 1) is widely active in the native African range of house and stable flies (Legner and Greathead 1969, Legner and Olton 1968). A new isolate from Israel possesses a high degree of drought resistance, aggressiveness and reproductive capacity that makes it a good candidate for filth fly control in the American west. Its ability to parasitize pupae at the filth habitat surface in cool weather especially suits it for *Fannia* control in California. Field data in southwestern California show that *Sphegigaster* sp. is capable of parasitizing such flies on poultry ranches, reducing their emergence by over 80% around release sites, with liberation rates of 100 per square-meter of breeding habitat.

A drought resistant strain of the pupal parasite *Spalangia endius* Walker from New Zealand (Legner and Olton 1968 and Legner unpublished data) also offers the possibility for a broader biological control in the semi-arid American west. This New Zealand relative also demonstrates activity at lower temperatures (22°C) than the resident form, and hybridizes with other strains of the same species where it is released. A population of hybrids can be produced which shows increased vigor over both parents in its ability to parasitize a greater number of flies within the same time period and in increased longevity. Commerical mass production is greatly facilitated because of the greater vigor and lower rearing temperature and humidity requirements.

A giant strain of *Muscidifurax zaraptor* Kogan and Legner (FIG. 2) occurs in the central part of the western Great Plains of Colorado and New Mexico (Legner 1977 and Legner unpublished data). This strain shows enhanced qualities of fecundity, cold-hardiness, heat tolerance and habitat foraging capability. Strains of this pupal parasite in California and the eastern United States are much more restricted in their ability to penetrate the breeding habitat in search of fly pupae, in their fecundities, and in their tolerance to high temperatures and winter cold.

A recent biological control success against stable flies, *Stomoxys* spp., on the Island of Mauritius has been achieved with the introduction of *Tachinaephagus stomoxcida* Subba Rao from Uganda (J. Monty, personal communication). Stable flies there had reached population densities of unprecedented proportions which plagued humans and animals alike (Monty 1972). The stable fly populations are now maintained at tolerable levels with annual releases of the parasite. Although a tropical species, *T. stomoxcida* offers possibilities of controlling stable flies in America through annual inoculative releases.

Although implementation of the above parasites is expected to improve biological fly control, abiotic and biotic requirements such as humidity, temperature, habitat depth and host preferences of the particular parasite species or strain must be considered to maximize their effectiveness (Ables 1975, Ables and Shepard 1976, Ables et al. 1976, Gerling and Legner

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FIGURE 1. New Israeli synanthropic fly parasite, *Sphegigaster* sp., examining house fly puparium prior to parasitization (photo by M. F. Badgley).

FIGURE 2. Giant cold hardy and heat resistant strain of *Muscidifurax zaraptor* Kogan & Legner from Colorado and New Mexico positioned on puparium of the common house fly (photo by M. E. Badgley).

1968, Legner 1977, Legner and Olton 1971, Legner et al. 1976, Markwick 1974, Mourier 1971, Olton and Legner 1974, Tingle and Mitchell 1975). Also, the delicate interrelationships that exist between parasitic insects and arthropod predators when their hosts are in balance at low densities (Legner 1969, Legner et al. 1980, 1981) demand a minimum disturbance of the breeding habitat to guarantee the continuance of this balance (Dietrick 1981, Legner 1981, Legner et al. 1973). Special considerations are required for different fly breeding habitats.

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ECOLOGICAL STUDIES OF INSECT PREDATORS AND *GAMBUSIA* IN RICE FIELDS: A PRELIMINARY REPORT

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ABSTRACT

Several experiments were conducted in rice fields in California's central valley during the 1981 rice season. The purpose of these experiments was to look at how the interaction between *Gambusia* and invertebrates may influence mosquito production. The preliminary results suggest that 1.) large predaceous insects reduce the abundance of small predaceous insects, 2.) *Gambusia* reduces the abundance and mean size of zooplankton, and 3.) insect predators prey more heavily upon *Culex tarsalis* in response to this reduced abundance of alternative food.

INTRODUCTION.–*Gambusia affinis* (the mosquitofish) consumes many prey types besides mosquitoes, and has been shown to alter the population densities of both zooplankton and predaceous insects (Reed and Hoy 1970, Farley and Younce 1977). It seems likely that these changes in the invertebrate community affect mosquito survival. Indeed, resurgences of mosquito numbers following the introduction of *Gambusia* to rice fields have coincided temporally with depressions in predaceous insect numbers (Hoy et al. 1972, Farley and Younce 1977). During the 1981 rice season we performed several experiments in rice fields, designed to investigate how *Gambusia* interacts with invertebrates to influence *Culex tarsalis* production.

METHODS.–We placed 24, 4 M^2 enclosures within a rice paddy during May 1981. One half of these enclosures were stocked with five adult female *Gambusia*. We attempted to remove predaceous insects from some of these enclosures by netting but found this to be not feasible, so we discontinued the netting after June 9th. Predaceous insects in the enclosures were sampled periodically by netting within a 1/4 M^2 aluminum quadrant. These insects were divided into three equal length classes: small, medium and large. Zooplankton and mosquitoes were sampled by taking 500 ml dips, using a glass jar.

A number of preliminary small-scale experiments indicated that the common predaceous insect species in rice fields reduced *Cx. tarsalis* survival (Bence in prep.). We therefore performed a short-term experiment that investigated the interaction between one of these predators, *Tropisternus lateralis* (a hydrophyllid beetle), and *Gambusia*. This two-way factorial experiment was done in 1/4 M^2 enclosures. All predaceous insects that could be caught were removed from 16 enclosures on July 10th, 1981. Thirty 2nd instar *T. lateralis* larvae were added to half of the enclosures. At this time *T. lateralis* was the numerically dominant predaceous insect and this was within the range of densities found in the field. Four large (>40 mm) *Gambusia* were then added to half of the enclosures with *T. lateralis* and half of those without *T. lateralis*, and then were removed after 2 days. One hundred fourth instar *Culex tarsalis* were then added to all enclosures. The number of *Cx. tarsalis* recovered by netting 12 h later was recorded.

The experiment was also an incomplete block design. Enclosures were placed in the field in pairs, and one member of each pair received *T. lateralis*, while the other did not.

RESULTS.–**Large Enclosures.** *Gambusia* reduced the density of zooplankton and shifted the size distribution of zooplankton towards smaller sizes. These samples have not been completely analyzed, however our data from July 25th illustrate this result (FIG. 1).

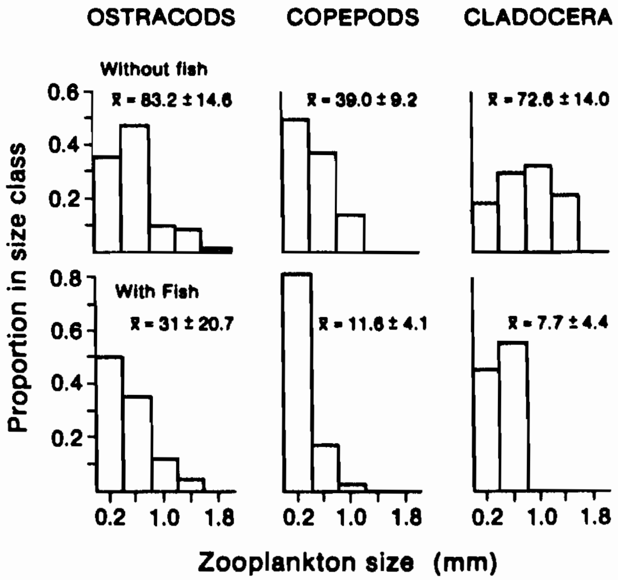

Figure 1. The size distribution of copepods, cladocera and ostracods in zooplankton samples from 4 M^2 enclosures with and without *Gambusia* on 7/25/81. The mean total number ± SE for each order is indicated on the figure.

Our attempts at removing all of the predaceous insects from the large enclosures failed. In fact, subsequent sampling suggested that there were more small predaceous insects in enclosures from which predaceous insects had been partially removed than from those in which predaceous insects were not removed (Table 1).

Small Enclosures. The results from the short-term small-enclosure experiment indicate that *T. lateralis* reduced the survival of *Cx. tarsalis*. A more unexpected result was that the presence of *Gambusia* prior to the introduction of *Cx. tarsalis* seemed to reduce *Cx. tarsalis* survival in those pairs of enclosures which contained *T. lateralis*, but not in those that did not (Table 2).

DISCUSSION.–Our results have shown that *Gambusia* reduces the abundance and mean size of zooplankton. In turn, results from our short-term small enclosure experiment indicate that *T. lateralis* may be a more effective predator of mosquitoes when alternative prey are scarce. (This is likely to be true of other predators also, including *Gambusia* itself.) Thus, one effect of *Gambusia* on predaceous insects is to increase their efficacy as mosquito predators by reducing the abundance of alternative prey.

Past work has shown, however, that *Gambusia* also reduces the abundance of predaceous insects (Reed and Hoy 1970, Farley and Younce 1977), and Bence (in prep.) has shown that some of these predator species reduce mosquito survival.

Table 1. Number of samples from large enclosures containing small predaceous insects (data pooled for 6-4-81 and 6-9-81).

	Predaceous Insects Removed	Predaceous Insects Not Removed
Small Predaceous Insects Present	20	8
Small Predaceous Insects Absent	22	30

ns	6-4-81	2X2	X^2= 1.5	$.1<p<.5$
*	6-9-81	2X2	X^2= 4.75	$.025<p<.05$
*	Summed	2X2	X^2= 6.25	$.025<p<.05$
*	Pooled	2X2	X^2= 6.17	$.01<p<.025$
ns	Heterogeneity		X^2=.08	$.5<p<.9$

Table 2. Number of *Cx. tarsalis* recovered from 1/4 M^2 enclosures after 12 hours.

WITH PREDACEOUS INSECTS			PREDACEOUS INSECTS REMOVED		
Block	*Gambusia* Present	*Gambusia* Absent	Block	*Gambusia* Present	*Gambusia* Absent
1	34	46	5	72	78
2	36	51	6	81	90
3	8	42	7	84	75
4	11	26	8	61	50

*	$F_{G(1,3)}$= 11.5 $.025<p<.05$	ns	$F_{G(1,3)}$= .017 $p>.75$
ns	$F_{blocks(3,3)}$= 4.5 $.1<p<.25$	*	$F_{blocks(3,3)}$= 10.5 $.025<p<.05$

* POOLED MANN WHITNEY $U_{(8,8)insects}$= 55 $p<.01$

In addition, by reducing the food supply of these predaceous insects, *Gambusia* may decrease their survival, growth and reproduction, while causing increased cannibalism and mutual predation among these predators. These effects reduce the efficacy of predaceous insects as control agents for mosquitoes.

Gambusia's own effects on mosquitoes probably are influenced by the abundance and size distribution of alternative prey, such as zooplankton. Furthermore, it seems that *Gambusia* on the one hand increases the efficacy of predaceous insects (by reducing alternative prey) but on the other hand reduces their efficacy by direct predation on the smaller instars (Bence and Murdoch in review) and by reducing their population density via reduction of the common food supply (zooplankton). Optimizing the combination of resident predaceous insects, and *Gambusia*, is not an easy problem. The results also suggest that effects of fish (and possibly of insecticides combined with fish) on non-target organisms may not always be detrimental to mosquito control.

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GAMBUSIA AS A PREDATOR UPON *NOTONECTA*: LABORATORY EXPERIMENTS

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ABSTRACT

Predation by *Gambusia* upon *Notonecta* has been suggested as a cause of resurgences in mosquito numbers, which have sometimes followed the introduction of *Gambusia*. We have conducted laboratory experiments that show that *Gambusia affinis* is a voracious predator upon the early instars of *Notonecta hoffmanni*. All sizes of fish that can consume more that one instar prefer the first instar. Additional experiments have shown that *Gambusia* will still consume *Notonecta* even when the mosquito *Culex pipiens quinquefasciatus* is present as an alternative prey. The number of *Notonecta* consumed depends upon the abundance of the alternative prey.

INTRODUCTION.–*Gambusia affinis* (the mosquitofish) preys upon many predaceous invertebrates and can sometimes reduce the abundance of these invertebrates, including that of *Notonecta* (Hoy et al. 1972, Farley and Younce 1977). Since *Notonecta*, the backswimming bug, is an important predator of mosquitoes (Hazelrigg 1974, Chesson 1981, Murdoch et al. in prep), we investigated the predatory interaction between *Notonecta hoffmanni* and *Gambusia* in the laboratory.

METHODS.–The experiments were conducted in eight liter aquaria. *Culex pipiens quinquefasciatus* and *Notonecta hoffmanni* were obtained from lab cultures, while *Gambusia* were either collected from the field or raised in the laboratory as necessary. There were six replicates in all cases. Controls (with no predators present) were conducted for all experiments.

For *Gambusia* predation experiments, three *Gambusia* of the appropriate size were introduced at least five days prior to the experiment and were not fed during the 24 hours preceding an experiment.

Each instar of *Notonecta* was exposed, separately (in groups of 40 for instars I-III and groups of 10 for instars IV and V), for two hours to large (>40 mm), medium (27-33 mm), small (17-23 mm) and very small (<10 mm) *Gambusia*. Only reasonable experiments were performed; for instance, medium-sized *Gambusia* could not eat *Notonecta* instar IV, and experiments using medium *Gambusia* and *Notonecta* instar V were therefore not performed. All four size classes of *Gambusia* were also presented with 10 *N. hoffmanni* eggs on a galvanized steel wire for at least 4 days.

Large and medium-sized *Gambusia* were exposed to mixtures of equal numbers of each *Notonecta* instar that they could consume, for 10 or 20 minutes respectively, to determine their preferred instar.

Medium and large *Gambusia* were presented with a mixture of 10 of their most preferred *Notonecta* instar (instar I) plus either 60 or 120, 3-4 mm *C. p. quinquefasciatus*. After 5 or 30 minutes for large and medium-sized *Gambusia*, respectively, the *Notonecta* were counted, the *Gambusia* and *Notonecta* removed, and the entire contents of the aquaria were strained through 250 micron Nitex netting and preserved in 95% ethanol.

Experiments investigating *Notonecta* predation upon *Gambusia* were performed in the same aquaria. The *Notonecta* were starved for 48 hours prior to the addition of very small *Gambusia*. A single *Notonecta* of instar IV, V or VI (adult) was used in each aquarium. The *Gambusia* were left for 64 hours after their addition, with occasional observations.

RESULTS.–No evidence was found of *G. affinis* predation upon *N. hoffmanni* eggs. Likewise, no evidence that *N. hoffmanni* prey upon *G. affinis* fry was seen.

Gambusia affinis is a voracious predator upon smaller instars of *Notonecta hoffmanni* (Table 1). Consumption rates for all sizes of *Gambusia* that fed upón more than one *N. hoffmanni* instar were greater for smaller instars. For any particular *N. hoffmanni* instar, larger fish consumed more during a two hour period.

Large *G. affinis* could consume *Notonecta* instar I-IV and showed a preference (by consuming more of this type when given equal numbers) for instar I in six our of six cases (FIG. 1). Medium-sized *G. affinis* could consume instars I and II and also preferred instar I in all six replicates (FIG. 1).

The rate at which *G. affinis* consumes instar I *N. hoffmanni* depends upon the abundance of mosquito larvae present. More *Notonecta* were consumed when fewer mosquito larvae were present (4.0 ± 0.3 and 7.2 ± 1.0 versus 1.0 ± .26 and 5.0 ± 0.9 for medium-sized and large *Gambusia*, respectively).

Table 1. Mean number of *Notonecta* remaining after two hours of exposure to different size classes of *Gambusia*. Note that initial numbers were 40 for *Notonecta* instars I-III and 10 for instars IV and V. Values are given plus or minus standard errors.

Notonecta instar	Control (No *Gambusia*)	*Gambusia* size Tiny (<10 mm)	Small (17-23 mm)	Medium (27-33 mm)	Large (>40mm)
I	39.5±.34	40	33.5±1.3	16.7±2.8	0
II	39.3±.33	-	39.7±.2	30.3±1.3	8.7±2.51
III	38.8±.48	-	-	39.7±.3	19.3±2.38
IV	10	-	-	-	8.0±.43
V	10	-	-	-	10

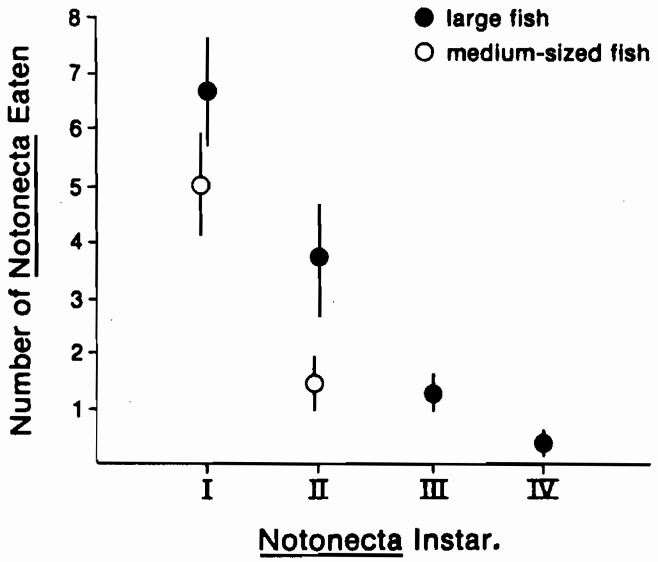

Figure 1. The mean number of each *Notonecta* instar eaten by large and medium-sized *Gambusia* when presented with equal numbers of instars I-IV or I and II respectively. The vertical bars indicate ± 1 SE.

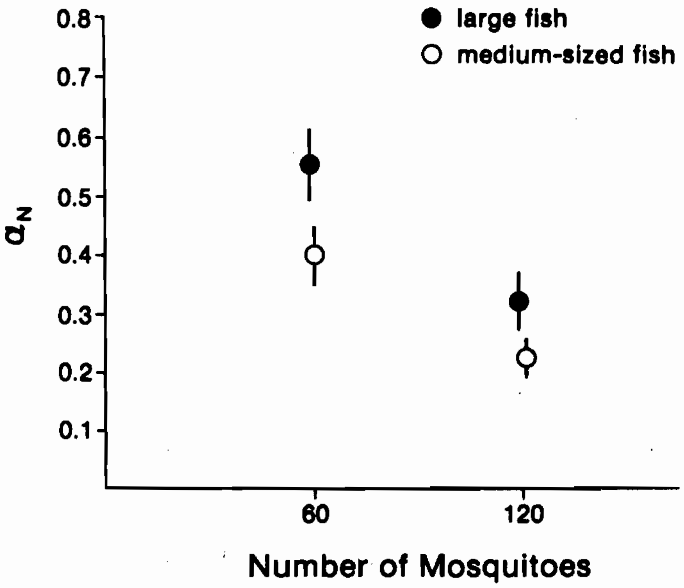

Figure 2. The mean preference (α) for *Notonecta* instar I of large and medium-sized *Gambusia* presented with 10 *Notonecta* instar I and either 60 or 120 *Culex pipiens quinquefasciatus*. The vertical bars indicate ± 1 SE.

This change appears to be greater than the proportionate change in relative abundances of the two prey species.

We tested this by calculating the preference statistic α_i using the formula:

$$\alpha_i = \ln \frac{R_i}{N_i} \Big/ \sum_{j=1}^{k} \ln \frac{R_j}{N_j} \qquad \text{(Chesson 1978).}$$

where R_i is the number of surviving prey, N_i the initial number of prey and k the number of different prey types. This form corrects for differences in relative abundance and is appropriate for cases (like ours) in which prey are depleted during an experiment. The sum of the α_i values is 1, equal values (1/k) indicate no deviation from random selection. Values of $\alpha_i > 1/k$ indicate preference for species i. Since in the two prey case α_i is approximately normally distributed, ANOVA was used to analyze the data.

The preference of *Gambusia* for *Notonecta* did depend upon the abundance of mosquito larvae and may also depend upon the size of fish. The results (FIG. 2) show a decrease in preference for *Notonecta* instar I when the number of mosquito larvae was doubled (while the number of *Notonecta* was held constant) ($p < .002$). At both relative densities the large fish appeared to have a higher preference for *Notonecta* than the medium-sized fish (FIG. 2), although the difference is not quite significant ($p = .06$).

DISCUSSION.—*Gambusia* may be an important predator of the early instars of *Notonecta*. However, within about two weeks at typical summer temperatures (Fox 1973), *Notonecta* reaches a size (instars III) at which the risk of predation by *Gambusia* is low, and eventually (instar IV) becomes essentially invulnerable to predation. Thus, the amount of predation by *Gambusia* upon a *Notonecta* population will depend on the density of larger *Gambusia* during periods when early instar *Notonecta* are present.

If alternative prey (including mosquitoes) are readily available when large *Gambusia* are abundant, *Notonecta* may still not be preyed upon strongly. *Gambusia*'s changing preference in relation to relative prey abundance indicates that they sometimes switch (in the sense of Murdoch 1969), and this behavior may further reduce predation on *Notonecta* when other prey are abundant.

Predaceous insect abundance, their size distribution and the abundance of alternative prey all change rapidly in temporary waters (such as rice fields) early in the season. Our results therefore suggest that the timing of *Gambusia* stocking in temporary water may be an important factor that determines whether a mosquito resurgence occurs.

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CURRENT DEVELOPMENT IN THE ENHANCEMENT OF MOSQUITOFISH CULTURE SYSTEMS

Robert L. Coykendall[1]

1. Fairly recent laboratory experimentation and field observations have demonstrated that the typical breeding season for mosquitofish populations of northern California can be dramatically extended by rearing them in waters at temperatures maintained constantly within the optimal breeding range of approximately 77° to 86°F (25° to 30°C). Thirty-fold annual increases in fish biomass have already achieved in geothermally-heated waters where this regimen has been provided.

We are now cooperating with the Klamath Vector Control District in developing a pilot fish production facility in southern Oregon. In addition to this geothermal project we are also communicating with a privately-owned power company about the potential conversion of one of their fossil fuel facilities to permit harnessing of what is now wasted heat. If this project is approved and funded, we may be able to develop a thermally-enhanced fish culture system for the northern Sacramento Valley.

2. We have long employed secondary municipal wastewaters in the rearing of our local fish, but often lose entire populations when effluent nutrient loading becomes excessive with respect to sudden adverse weather and/or seasonal climatic changes. We have applied for and received permits from appropriate State and local agencies to construct rearing ponds adjacent to an existing facility and we plan to utilize its secondary effluent as a water and nutrient source. This will be beneficial as we should be able to better manage effluents, yet achieve high growth rates with minimal supplemental feeding and fertilization.

3. At our own headquarters we are currently in the process of designing a small intensive culture system utilizing a solar greenhouse with will enclose large, yet low cost permanent tanks.

4. Finally, for the last few years we have cooperated with the aquatic weed control specialists at University of California at Davis in the experimental stocking of our mosquitofish ponds with a certain variety of common carp– the mirror carp. This fish with its rooting behavior has significantly inhibited aquatic weed and filamentous algal growth to the extent that normally-required chemical applications to allow fish harvests have been reduced or eliminated. More importantly, last year's study indicated that there was little or no observable predation upon our mosquitofish stocks. Current studies are being conducted to further assess any possible adverse effects and to determine appropriate stocking rates for the carp in our mosquitofish ponds.

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APPLICATION OF A CAPTURE - RECAPTURE METHOD FOR ESTIMATING DENSITIES OF MOSQUITOFISH POPULATION IN RICE FIELDS

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ABSTRACT

A simple method to estimate absolute density of mosquitofish population in rice fields was described. It is still at a preliminary stage, but this method allows us to predict absolute abundance from relative abundance data (average number mosquitofish captured/trap).

INTRODUCTION.–In making integrated pest management (IPM) models, accurate assessment of densities of pests and natural enemies in their ecosystems must be obtained. Capture recapture methods provide a means of estimating the density of populations and are also useful in understanding the dispersal behavior of the species being studied.

Marking methods used of average sized fishes such as, tagging, fin-clipping, branding, injection, immersion, feeding, daubing and tatooing are greatly limited with mosquitofish because of their small size. O'Grady and Hoy (1972) successfully used fluorescent dye (Rhodamine B) to mark mosquitofish for short-time population studies. A preliminary study of mass marking of mosquitofish by using fluorescent polystyrene pigment forced into the dermal tissue with compressed air was reported by Vondracek et al. (1980).

In 1981 at the Fresno laboratory, we began investigating the feasibility of mass marking mosquitofish with polystyrene pigments to use in studying about their dispersal and population dynamics.

METHODS AND MATERIALS.–Mosquitofish used for this study were collected from Koda's rice field number four. They were maintained in a metal tank at a water temperature of ca. 22°C, 16L:8D photoperiod and 60% sat. dissolved O_2. Detailed marking procedures used were reported by Stewart and Miura (1982), but in brief, fish were marked by spraying them with red fluorescent pigment under high pressure (175 psi) for 10 seconds then immediately washed with water and returned to a holding tank containing treated water (2 ppm $KMnO_4$ and 500 ppm NaCl) and held for several days before release.

A viewing box equipped with a glass aquarium and black light was constructed and used for detecting marked fish in fields.

The north end of a single paddy (ca. 1 acre size) of Koda's rice field number four was used for this study. Prior to release, the weir boxes (inflow and outflow) were screened to prevent fish movement into and out of the paddy. Eight hundred marked fish were released on July 27, 1981. The paddy was arbitrarily divided into five sections and five minnow traps were randomly set in each section and left for 24h. All captured fish were examined under the black light for the red marking and then released in the same section where they were captured. Numbers of marked and unmarked fish captured for each section were tabulated.

Absolute population size at each trapping time was calculated by using the Lincoln Index (Southwood 1978):

$$N = \frac{M \times C}{m}$$

where N = the population size in the study area, M = total number marked fish released, C = total number fish captured, m = total number mark fish recaptured.

RESULTS AND DISCUSSION.– Table 1 shows results of the capture-recapture study. The average number mosquitofish recaptured was ca. 4% of the total number captured; day 3 was an exception as 17% of the marked fish were recaptured, indicating that the marked fish had not yet dispersed in the paddy. The estimate of absolute population using the Lincoln Index and relative abundance of fish in the paddy are also shown in Table 1.

· The relationship between the two variables (absolute and relative abundances) is shown in Figure 1. There is a significant positive correlation between the variables (r = 0.9446), i.e. as the absolute population density of mosquitofish increases, the number fish collected/trap also increases. In order to predict absolute density of population from trap data (average number fish collected/trap), the collection data were analyzed using regression analysis (Figure 1) and a linear regression equation was calculated as $Y_i = 0.83 + 0.0011X_i$ with SE of intercept = 2.89. By using this equation, it is easy to estimate absolute density of mosquitofish from any

TABLE 1. Absolute population estimation of mosquitofish by the capture-recapture method[a].

Day of capture	No. fish/ trap	No. marked fish	No. unmarked fish	Estimated no.[b] fish/acre
3	7.32	31 (17.0)[c]	152	4722
15	15.20	19 (5.0)	361	16000
17	27.56	23 (3.3)	666	23967
18	23.36	25 (4.5)	559	18688
21	13.32	18 (5.4)	315	14800
25	24.52	23 (3.8)	590	21321
30	21.92	20 (3.6)	528	21920

[a] 800 marked fish (avg. 3-4 cm fork length) were released on July 27, 1981.

[b] No. fish/acre = total no. marked fish released x total no. fish captured ÷ total no. marked fish captured.

[c] Numbers in parenthesis are % captured.

TABLE 2. Estimated numbers of mosquitofish for small collections (<15 fish /trap)[a].

No. fish /trap	No. fish /acre	No. fish /ha	No. fish /trap	No. fish /acre	No. fish /ha
0.1	91	225	4	3312	8181
.2	182	450	5	3980	9831
.3	273	674	6	4889	12076
.4	364	899	7	5898	14568
.5	454	1121	8	6896	17033
.6	545	1346	9	7805	19278
.7	636	1571	10	8714	21524
.8	727	1796	11	9622	23766
.9	818	2020	12	10532	26014
1.0	909	2245	13	11441	28259
2.0	1688	4169	14	12350	30504
3.0	2500	6175	15	12882	31818

[a] 2 regression lines, $Y = 0.83 + 0.0011X$, and $Y = 0.001X$ were drawn and values of Xs' were adjusted by graphically.

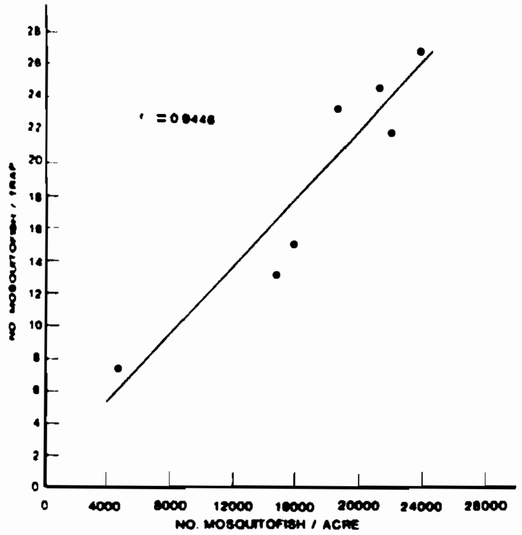

Figure 1. Scatter diagram for data in Table 1 and a regression line of relative abundance on absolute abundance (no. fish/acre).

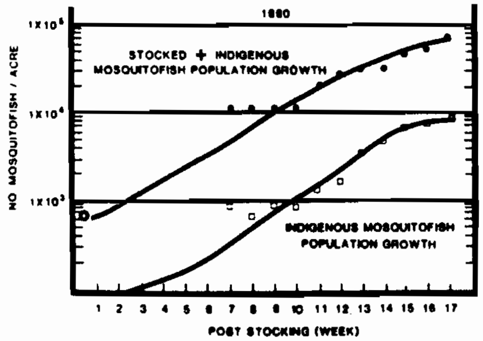

Figure 2. Population dynamics of mosquitofish in rice fields in 1980. Initially 600-700 mosquitofish were stocked per acre in May.

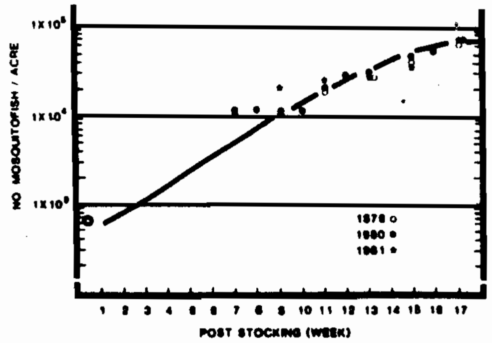

Figure 3. Comparison of three years population dynamics of mosquitofish in Fresno County rice fields (initial stocking = 600-700 fish/acre).

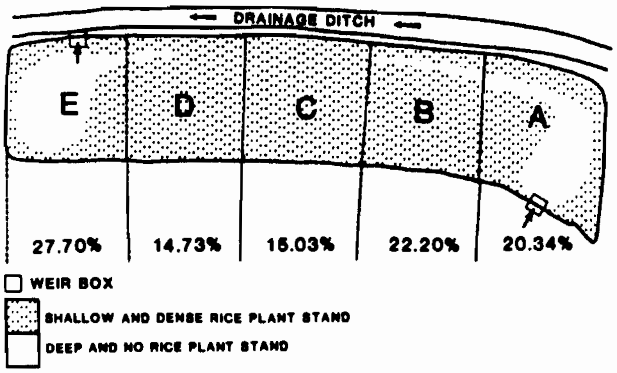

Figure 4. Schematic diagram of the paddy used to study a capture-recapture method, showing physical condition and distribution of mosquitofish. Arrows indicate general water movement.

trap datum. However, the regression line did not intercept at the origin (if the two variables are directly proportional, the line should intercept at the origin) special table values for smaller trap collections (<15/trap) were calculated to make better estimations of absolute population sizes and are shown in Table 2.

In order to test applicability of this method, the Koda's 1980 field collection data were transformed into absolute abundance and are shown in Figure 2. Initially, ca. 600-700 mosquitofish/acre were stocked in this field in early May by the Fresno Westside MAD. By late June the population density had grown to the 10,000 level and at the end of August (17 weeks post stocking) it reached almost 100,000 fish/acre. Mosquitofish are well established in the Fresno Westside rice growing area, in an adjacent unstocked field containing only an indigenous population of mosquitofish, ca. 10,000 fish/acre were estimated at the end of August.

Our relative abundance data of 1980, 1981 and those of Reed and Bryant (1975) are also transformed into the absolute abundance and shown in Figure 3. The estimated numbers for each datum are surprisingly in a good agreement with each other.

This method is of value in understanding the dispersal and spacial distribution behavior of mosquitofish in rice fields. Figure 4 illustrates physical characteristics of the paddy and the distribution of fish within. About 48% of fish were aggregated in deep areas where rice stands are scant, while the traps in the shallow water and dense rice stands (Section C and D) contained fewer fish. Directional flow of water in the paddy probably does not effect the fish distribution, ca. equal number of fish were present in sections A + B and D + E. Those findings are generally in agreement with the results reported by Reed and Bryant (1972), Norland and Bowman (1976) and Miura et al. (1979).

In summary, it is still at a preliminary study but the main point to show is that even relative abundance data can be used to estimate absolute density by using this method.

ACKNOWLEDGMENT.–This study was funded, in part, by a special California State appropriation for mosquito control research, and by EPA cooperative agreement No. CR-806771-01-1.

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A PRELIMINARY REPORT ON THE USE OF MOSQUITOFISH TO CONTROL MOSQUITOES IN AN URBAN STORM DRAIN SYSTEM

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INTRODUCTION.–Underground storm drain systems have been constructed in most urban areas of California as a means for transferring runoff water from city streets to impoundment basins. For various reasons, the design of many storm drain lines is such that some water is trapped within the underground system. These underground areas are capable of producing large numbers of *Culex quinquefasciatus* Say, which are difficult to control because of the very limited access to the mosquito breeding areas (Hazelrigg and Pelsue 1980. Mulligan and Schaefer 1982).

Mulligan and Schaefer (1982) described an effective method of physically preventing access of the mosquito to the underground system. With the limitations of chemical control methods in storm drains, there is a need for further study in physical and biological control strategies. The following is a preliminary report on the use of mosquitofish *Gambusia affinis* to control mosquitoes in storm drains.

MATERIALS AND METHODS.–The Jackson storm drain line was selected for study. Located approximately one mile east of the 7th Street line studied by Mulligan and Schaefer (1981, 1982), it is composed of 3.25 miles of various sized lateral lines which connect to a 1.75 mile long, 54 inch diameter trunk line. The trunk line empties through an outflow structure into a ponding basin located at the Fresno District Fair Grounds.

The trunk line is designed as a "zero-grade" line meaning it is constructed with essentially no net difference in elevation between the origin of the line and it's termination at the outflow structure. Due to construction problems, however, the line is composed of sections with either slightly positive or negative slopes. There is a vertical displacement of the line in a manhole chamber approximately 0.6 miles upstream from the outflow structure. The outlet line is approximately one foot higher than the inlet line which results in water backing up in the upper 1.15 mile of trunk line.

Ten pounds of unsorted mosquitofish were stocked into the upper one-half mile of the trunk line on June 5, 1981. The fish were divided equally among four manhole chambers (1-4 on figure 1). Approximately five pounds of mosquitofish were stocked in lateral lines (manhole chambers 5-9) on June 23, 1982. All fish were obtained from local sources.

The occurrence of fish in unstocked portions of the line was determined by removing the manhole cover and visually inspecting the manhole chamber. The fish were then captured by dip net and taken to the lab for dissection to determine gut contents and reproductive status.

Miniature CDC-type light traps were placed in manhole chambers 10-11 (0.7 and 0.8 miles upstream from the outflow structure). The traps were loaded with approximately 2 lbs. of dry ice and operated overnight for one to five nights

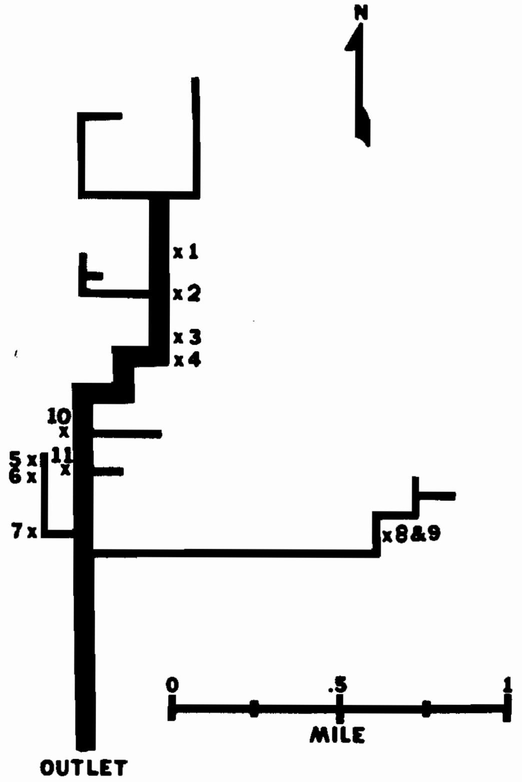

Figure 1. Diagram of the Jackson storm drain line. (xN) denotes manhole chamber N. The heavy line is the trunk line, thinner lines represent laterals.

	Jackson Line		7th Street Line	
	Adult Mosquitoes Per Trap Night		Adult Mosquitoes Per Trap Night	
	1980	1981	1980	1981
May	-	62	-	1555
June	-	2148[1]	-	4050
July	2156	283[2]	2625	2667[4]
August	2737	66	2588[3]	1695
September	--	7	822	207
October	-	3	1132	-

1. Fish stocked in Jackson Line June 5, 1981.
2. Fish found throughout Jackson Line July 15, 1981.
3. Treatments with Malathion by Mistblower August 15 & 28, 1980. Treatment with Cythion by Coldfogger September 12, 1980.
4. Physical control devices installed by U.C. Mosquito Research Lab July 27-28, 1981.

Figure 2. CDC-type light trap counts for the Jackson and 7th Street storm drain lines.

per month by personnel of the U.C. Mosquito Research Lab in Fresno who also provided the 1980 mosquito data for the Jackson line and the 1980 and 1981 data for the 7th Street line.

RESULTS AND DISCUSSION.–Mosquitofish survived the nearly lightless conditions in the storm drain throughout the summer and established themselves throughout the trunk line within six weeks of stocking. With the establishment of the fish, the light trap counts of adult *Culex quinquefasciatus* Say began a dramatic decline (figure 2). A comparison of trap counts between 1980 and 1981 in the Jackson line shows that the June 1981 counts were in the same range as the July and August 1980 counts. The mosquitofish were stocked upstream from the section of line monitored by light traps but had migrated into the light trap area by July. The light trap counts dropped from a mean of 2148 mosquitoes per trap night in June to a mean of 283 mosquitoes per trap night in July. The counts continued to drop through the end of the study in October. By contrast the 1981 mosquito counts in the 7th street line remained at approximately 1980 rates through July and then began to drop in August after the physical control barriers were installed.

The physical condition of the fish in the stocking area began to decline several weeks after stocking. The emaciated condition was probably due to starvation caused by overstocking and subsequent over-utilization of environment. Fish which had migrated downstream were in markedly better condition, the smaller numbers utilizing the environment more conservatively.

Gut samples indicated that in addition the mosquito larvae, the fish were preying on snails, copepods, annelids, psychodids and miscellaneous terrestrial invertebrates which were washed into the line. As the season progressed the fish became more dependent on terrestrial organisms as the aquatic fauna were decimated.

Female mosquitofish contained eyed egg stages when sampled two weeks after stocking and fry were found in the line up to six weeks after stocking. However, after six weeks no further evidence of reproduction was found. Females caught after six weeks contained no developing embryos, indicating that no mating was taking place in the storm drains.

Though these results are preliminary and circumstantial in some cases, we feel that the use of mosquitofish to control mosquitoes in storm drains holds great promise because they survived in the storm drain for at least four months, they fed upon mosquitoes, and they survived on alternate prey as the number of mosquito larvae was reduced.

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STUDIES ON THE RELATIONSHIP OF MOSQUITO BREEDING ON RICE FIELDS AND THE USE OF SEWAGE EFFLUENT FOR IRRIGATION

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ABSTRACT

The use of sewage effluent for irrigating rice fields in the San Joaquin Valley, results in the production of high populations of *Culex tarsalis*. These populations cannot be controlled effectively by existing technology and no improved methods appear to be forthcoming. Therefore all legal recourses should be used by public agencies to eliminate the practice of discharging sewage effluent onto San Joaquin Valley rice fields.

INTRODUCTION.–The breeding of *Culex* spp. mosquitoes in the water of sewage and dairy-waste holding ponds is well known and constitutes a serious problem in Central California. The breeding potential in such habitats can be adequately reduced by maintaining steep banks, controlling emerged vegetation and, in those ponds not highly-polluted with organic debris, by stocking with mosquitofish. In such ponds, if the bank steepness deteriorates and shallow water areas result, there is a rapid growth of weeds and such conditions provide optimum breeding sources for *Culex* mosquitoes; this necessitates the application of chemicals and over a long period of time has led to the development of intense insecticide-resistance in the San Joaquin Valley.

During the past three years the Mosquito Control Research Laboratory has contracted with the U.S. Environmental Protection Agency (through Texas A & M University) to study the mosquito breeding problem in California rice fields. The main objective of this research is to determine the impact of rice culture practices on mosquito breeding. Study fields have been selected in Fresno, Kern and Merced Counties; the extent of mosquito breeding was measured by quantitative sampling procedures. On rice fields in Fresno and Kern Counties which (1) are irrigated with canal water, (2) on which weeds are controlled and (3) which are stocked with mosquitofish, mosquito breeding generally averages less than 0.1 larvae plus pupae per dipper sample. Chemical control is generally not required on such fields. During 1980 a rice field in Merced County, which was irrigated with a mixture of canal water and sewage effluent, produced large populations of *Culex tarsalis* (average 3-4 larvae + pupae per dip sample. The level of mosquito production was ca. 30X greater than for fields simultaneously being monitored, but where only fresh irrigation water was used. During 1980 approximately 500 acres of rice was grown on fields owned by the City of Bakersfield and leased to a farmer which were irrigated with water containing sewage effluent. Large numbers of *Culex tarsalis*, as well as lower numbers of *Culex quinquefasciatus*, were produced; control efforts by the Kern Mosquito Abatement District did not achieve satisfactory results. These difficulties were caused by the presence of a heavy algal growth which prevented spray penetration and which restricted the effective predation by mosquitofish and because of insecticide-resistance in the mosquito populations. During 1981, research studies were initiated to define the relationship between mosquito breeding in rice fields and the use of sewage discharges in the irrigation water.

MATERIALS AND METHODS.–Two areas were selected for research studies during 1981: (1) a rice producing area of Fresno County located northwest of Firebaugh where this crop has been consistently grown for many years and where management practices are representative of those generally used in the San Joaquin Valley (use of canal water and stocking with mosquitofish) and (2) rice fields on properties of the City of Bakersfield being farmed by a grower who uses a mixture of treated sewage effluent and fresh water; the latter fields were also stocked with mosquitofish.

In each area, one field was selected for intensive monitoring throughout the rice irrigation season. In Fresno County, a 20 acre field (Koda rice field No. 4) was selected as being representative of the management practices usually employed. In Kern County a 40 acre field (Garone rice field No. 5), which was being irrigated with water containing sewage effluent, was selected for comparison; this field was divided by a north-south border into two subparts which were designated 5E (east portion) and 5W (west portion) and these were approximately 20 acres each.

On each field, mosquito immatures and members of the predator complex were sampled bi-weekly. The sampling was alternated so that one field was sampled during a given week and the other on the consecutive week. Mosquito immatures were collected by standarized dipping procedures along defined transects and the predators by dipping, trapping and by area sampling. Samples of mosquito larvae collected in the field were also taken to the Fresno Laboratory to confirm species identification.

The sampling plan at the Garone rice field area had to be modified as the grower shifted to the practice of intermittent irrigation, which was not anticipated. The irrigation practice he used was to flood for 10-14 days followed by a drying cycle of ca. seven days. Since Garone rice field No. 5 was dry on some of the planned sampling intervals, other rice fields in the same area and being managed in the same general pattern, were also sampled for immature mosquitoes; these included fields No. 74A and No. 78A.

In addition an attempt was made to check with mosquito

TABLE 1. Numbers of immature Culex tarsalis collected from Garone rice field no. 5 during 1981.

Date (1981)	Field	Immature I	 II	 III	 IV	 P	Total no.	No. dips	Mean no./dip
7/ 7	5-West	21	18	13	9	0	61	40	1.525
	5-East	8	48	16	14	1	87	40	2.175
7/21	5-West	0	4	3	0	0	7	20[a]	0.350
	5-East	0	1	0	0	0	1	19	0.053
8/18	5-West	0	9	8	0	0	17	17	1.000
	5-East	17	24	8	6	0	45	18	2.500
9/ 1	5-West	0	0	0	0	0	0	23	0.000
	5-East	0	0	0	0	0	0	20	0.000
9/ 9	5-West	0	3	7	5	1	16	39	0.410
	5-East	0	1	18	2	0	20	16	1.312

[a] Less than 40 samples indicates that part of the field was dry.

TABLE 3. Numbers of immature Culex tarsalis collected from Koda rice field no. 4 during 1981.

Date (1981)	Immature stage I	 II	 III	 IV	 P	Total no.	No. dips	Mean no./dip
6/30	5	4	5	1	1	16	52	0.31
7/14	0	0	0	1	0	1	52	0.02
7/28	0	0	0	0	1	1	52	0.02
8/11	0	2	0	0	0	2	52	0.04
8/25	0	0	0	0	0	0	52	0.00

abatement agencies to (1) find other situations where irrigation water containing sewage effluent was being placed on rice and (2) to compare mosquito production on the Garone rice fields with that of other rice fields in the same general area where only fresh water was being used for irrigation.

RESULTS AND DISCUSSION.–The numbers of immature *Culex tarsalis* produced on the Garone rice field No. 5 during 1981 are shown in Table 1. These represent very high levels of breeding (note: some mosquito abatement districts treat when the immature population density reaches a mean of 0.1 per dipper sample). On two sampling dates (7/21 and 9/1) the numbers were lower but this was due to the fact that the water had recently been applied to the field–note that only early stage immatures were present on these dates. As a result of the intermittent irrigation, the mosquitofish populations were killed during the drying phase, yet the water still persisted long enough to allow a large number of the immature mosquitoes, which were present, to complete their life cycle.

On 8/5/81 the Garone rice field No. 5 was dry, so sampling was conducted on Garone rice fields 74A and 78A. Table 2 shows the numbers present on that date. The water on field 78A was much more recent as shown by the presence of smaller stage immatures but the extent of breeding represents an intolerable level.

For comparison the numbers of immature *Culex tarsalis* produced on the Koda rice field No. 4 are shown in Table 3; it was only during the early season (6/30), prior to the build-up of the predator population, that a significant breeding problem existed. Such results are similar to previous findings in Kern County (Schaefer et al. 1981).

Comparisons of the aquatic organisms occurring on these rice fields, as determined by three separate sampling methods, are given in Tables 4, 5 and 6. Mosquitofish are most effectively collected by trapping (Table 5) and their seasonal build-up at the Koda rice field No. 4, following a single stocking, is clearly shown; in contrast, multiple stockings of mosquitofish at high densities on Garone rice field No. 5 did not allow a seasonal build-up of the population because of the drying cycle which killed them. The area samples comprise the water column and the upper mud layer and show that much larger numbers of midges developed on Garone rice field No. 5 than at Koda rice field No. 4. These midges create a public nuisance problem but they are not disease vectors.

Figure 1 gives a seasonal comparison of the populations of immature mosquitoes and of mosquitofish on Garone rice field No. 5 and Koda rice field No. 4.

The breeding levels on the Garone rice fields were such that frequent aerial applications against both larvae and adults were made by the Kern MAD; this amounted to treating a cumulative total of 5,710 acres. Since there were 1,100 acres of rice the required treatments were 519%. Only partial control was obtained and additional insecticide treatments by ground equipment were also made (Yoshimura 1981).

Another farmer in an area adjacent to the Garone rice fields grew 634 acres of rice but used only canal water for irrigation.

TABLE 2. Number of <u>Culex tarsalis</u> immatures on Kern County rice fields on August 5, 1981.

Field	No. Transect	Immature Stage I	II	III	IV	P	Total	N[b]	Average no./sample
74A	A	3	7	6	13	7	36	20	1.800
	B	1	0	0	0	0	1	19[a]	0.053
	C	0	18	1	0	0	19	20	0.950
	D	39	61	45	37	21	203	17	11.941
total		43	86	52	50	28	259	76	3.408
78A	A	0	0	0	0	0		20	
	B	0	0	0	0	0		20	
	C	0	0	2	0	0	2	20	0.100
	D	18	3	0	0	0	21	20	1.050
total		18	3	2	0	0	23	80	0.288

[a] Less than 20 samples per transect indicates a portion of the field was dry.

[b] No. of samples.

TABLE 4. Numbers of aquatic organisms collected from rice fields in 40 dips during 1981.

Organism	Garone rice field no. 5E						Koda rice field no. 4				
	Intermittently irrigated with well and sewage water						Continuously irrigated with canal water				
	July		Aug.		Sept.		June	July		Aug.	
	7	21	5	19	1	8	30	14	28	11	25
Water flea	4	3	Dry	104	621	2626	212	315	454	850	813
Copepod	2881	2995	"	2882	3583	4056	411	295	303	628	1360
Seed Shrimp	37	59	"	39	47	84	1575	2181	2491	3625	3921
Mayfly	36	34	"	44	2	0	4	5	13	7	4
Damselfly	16	22	"	13	1	1	62	41	52	67	54
Dragonfly	0	0	"	22	6	2	6	1	3	2	0
Beetle adult	0	1	"	0	0	0	0	1	0	0	0
Beetle larva	8	10	"	26	5	3	7	4	2	3	1
Midge	85	90	"	365	145	476	34	15	11	109	111
Biting midge	6	2	"	3	4	4	2	5	12	6	0
Shorefly	11	14	"	23	7	4	2	4	0	5	0
Mosquito larvae	61	2	"	57	2	328	11	4	4	3	2

TABLE 5. Numbers of aquatic organisms collected overnight in 24 minnow traps during 1981.

Organism	Garone rice field no. 5E					Koda rice field no. 4				
	Intermittently irrigated with well and sewage water					Continuously irrigated with canal water				
	July		Aug.		Sept.	June	July		Aug.	
	7	21	5	19	1	30	14	28	11	25
Damselfly	12	0	Dry	2	1	43	42	35	43	21
Dragonfly	1223	81	"	16	0	18	19	19	30	21
Water boatman	42	4	"	30	10	14	0	0	1	2
Backswimmer	49	10	"	3	4	22	3	0	0	0
G. water bug	3	4	"	40	6	9	10	17	7	3
Beetle adult	1705	682	"	1256	482	182	281	105	108	21
Beetle larva	2	161	"	185	69	8	8	13	22	9
Mosquitofish	2412	2588	"	3	3	572	649	734	832	1772

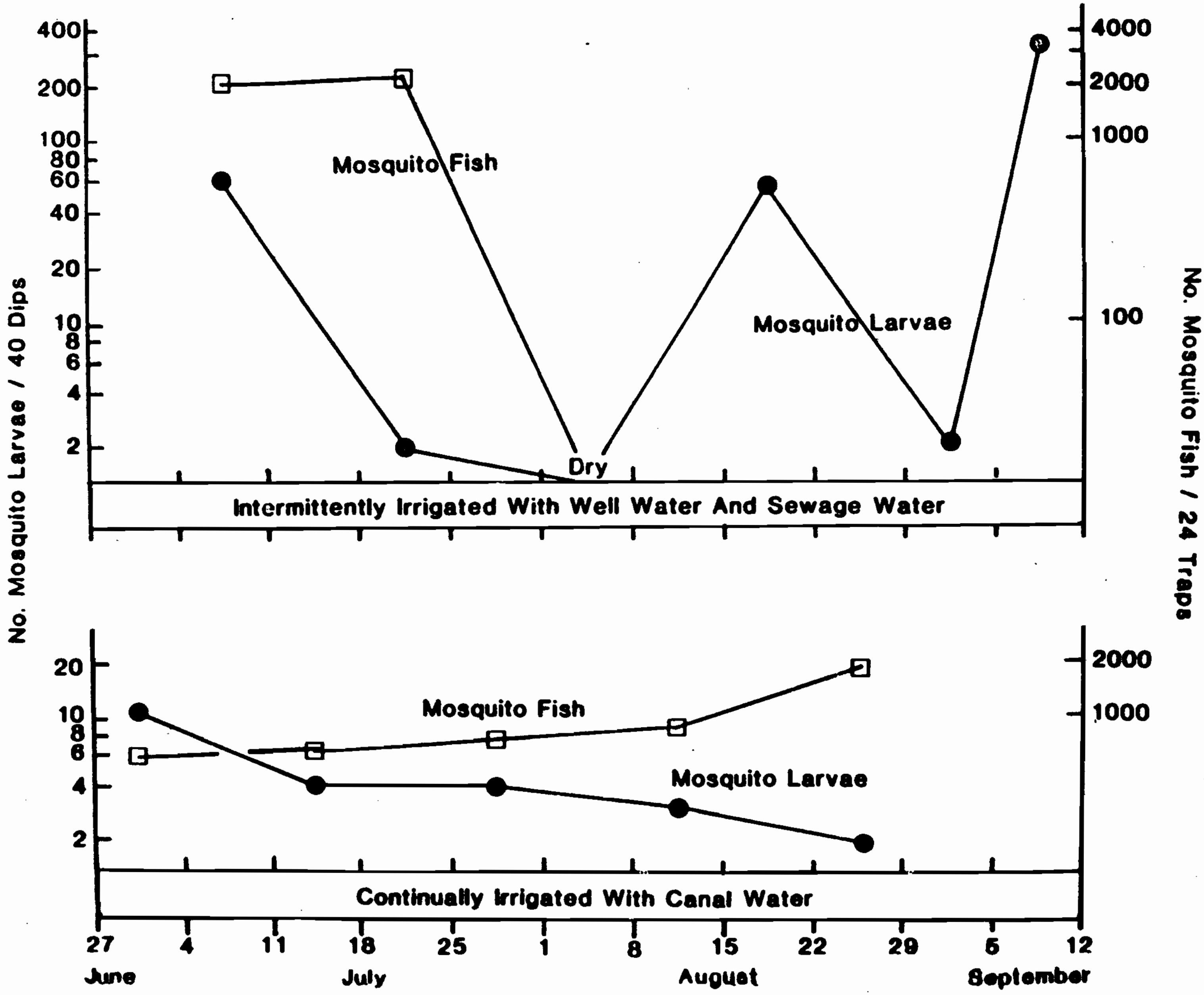

Figure 1. Comparision of populations of mosquitoes and mosquitofish on Garone rice field No. 5 and Koda rice field No. 4 during 1981.

TABLE 6. Numbers of aquatic organisms collected in 26 area samples from rice fields during 1981.

Organism	Garone rice field no. 5E Intermittently irrigated with well and sewage water July 7	21	Aug. 5	19	Sept. 1	Koda rice field no. 4 Continuously irrigated with canal water June 30	July 14	28	Aug. 11	25
Mayfly	587	1	Dry	0	2	40	14	16	73	73
Damselfly	180	68	"	0	0	1183	1261	1194	1474	1235
Dragonfly	79	34	"	55	0	40	68	62	122	99
Beetle adult	25	14	"	63	40	37	5	10	20	8
Beetle larva	134	54	"	59	32	64	54	29	37	12
Midge	8974	8813	"	3417	2376	125	49	26	508	1437
Biting midge	162	67	"	123	24	10	58	252	181	470
Shorefly	341	142	"	81	112	1	3	15	41	51
Other fly	29	33	"	2	9	32	65	182	125	80
Mosquito	54	0	"	50	6	3	7	3	3	7
Mosquitofish	19	19	"	0	0	4	7	13	10	33

These fields were also stocked with mosquitofish. Some insecticide applications by the Kern MAD were required prior to the time that the mosquitofish populations became established and where bermuda grass grew on the borders. Only 15.6% of the total rice area required chemical treatments (Yoshimura 1981).

During 1981, we learned that a 640 acre block of rice in Kings County was being irrigated with canal water into which sewage effluent had been discharged. The Kings MAD informed us that this was their worst source of breeding of *Culex tarsalis* and that breeding in fields irrigated with water containing sewage effluent was much greater than on other rice fields in that area where only fresh water was used (Dawson 1981). Zaim and Newson (1980) described the effects of water containing sewage effluent on the mosquito breeding problem; they summarized, "Observations to this time clearly indicate that the disposal of sewage effluent by spray irrigation creates a diversity of excellent mosquito breeding habitats and the resulting increases in the populations and species diversities enchance the potential for pest and disease transmission problems in the vicinity of this and other similar sewage disposal projects. Planners, designers, and operators of these types of sewage disposal systems need to be fully aware of the potential problems inherent in these types of sewage disposal systems in order that they can incorporate appropriate safeguards in their design and operations. Otherwise, the widespread adoption of sewage disposal by land irrigation may resolve immediate problems but create new health hazards for the communities in which they are located."

CONCLUSION.–*Culex tarsalis* production on rice fields which were irrigated with water containing sewage effluent and were either flooded continuously, e.g. Merced and Kern Counties during 1980 and in Kings County during 1981, or were intermittently flooded, e.g. Kern County during 1981, clearly shows the massive breeding potential under these conditions. This practice results in not only a public health nuisance, but since *Culex tarsalis* is the major vector of both St. Louis and western equine encephalitis in this region, and since the virus causing these diseases is also being reported in the same region, there is a very high risk of an outbreak of human encephalitis. The use of sewage effluent on rice fields promotes algal growth which forms mats which restrict spray penetration and which limit the mobility, and thereby the effectiveness, of mosquitofish. In addition, the severe problem of insecticide-resistance in the local populations of *Culex tarsalis* makes them "immune" to commercially available insecticides. No new technology to solve such a problem appears to be forthcoming in the near future. This results in a situation where mosquito abatement districts are unable to cope with this problem in an effective manner. Because of the resulting health hazard potential, sewage effluent should not be discharged onto rice fields.

RECOMMENDATION.–All legal recourses by public agencies should be used, as necessary, to eliminate the practice of discharging sewage effluent onto rice fields in the San Joaquin Valley. Failure to take such action may result in a public health catastrophe of the type that occurred in Kern County in 1952; this event resulted in not only loss of many human lives but also in the permanent brain damage of young children, some of which are still institutionalized as permanent wards of the State (Finely et al. 1967). It would be very foolish and irresponsible to presume that another epidemic will not occur and continue to allow high densities of this vector species to prevail until the local population is subjected to a major disease outbreak.

ACKNOWLEDGEMENT.–The cooperation of personnel of the Fresno Westside, Kern and Kings Mosquito Abatement Districts is gratefully acknowledged. This research was funded, in part, by EPA Cooperative Agreement No. CR-806771-01-1 and by the Kern Mosquito Abatement District.

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MOSQUITO OCCURRENCE IN WASTEWATER MARSHES: A POTENTIAL NEW COMMUNITY PROBLEM

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THE WASTEWATER MARSH CONCEPT.–In California there is increased interest in developing wastewater marshes as an alternative method for improving wastewater treatment. The State Water Quality Control Regulations require that sewage effluent must meet advanced secondary or tertiary treatment requirements before being discharged into waters of the state (streams, rivers, estuarine areas and bays).

Where sufficient land is available, the use of a marsh functioning as a "living filter" to lower BOD and reduce levels of coliform and nutrients from wastewater would cost considerably less than a conventional high technology tertiary treatment facility would cost. There would also be a substantial reduction in energy cost by using a marsh system. The marsh system is receiving greater attention since federal funds for wastewater treament facilities are being curtailed.

Other beneficial uses cited for using the marsh system for wastewater treatment are (1) increased acreage of wetland habitat for wildlife enhancement to provide for conservation of wildlife, (2) educational purposes and (3) recreational needs. In estuarine brackish marsh areas the addition of fresh wastewater aids in lowering salinity levels and improves the diversity of vegetation for waterfowl food.

In California the State Senate Resolution SR 28, September 1979, has called for an increase in State wetlands by 50% by the year 2000. Sheehan (1980) reports the present acreage of wetlands at 500,000 acres and the projected increase would add 250,000 acres in 20 years. The key limiting factor in the expansion of wetlands is availability of water. The State Water Resources Control Board estimates that the use of treated municipal wastewater could supply about 10% of the demand for future wetland requirements. From a national viewpoint, EPA issued a notice of intent in the Federal Register, Vol. 46, December 18,1981, for the proposed disposal of wastewater to freshwater wetlands in eight southwestern states.

THE IMPACT OF WASTEWATER MARSH SYSTEMS ON MOSQUITO PRODUCTION.–The emphasis on wastewater marsh systems creates new conditions near municipalities that can provide ideal habitats for mosquito propagation unless these systems are properly designed and managed. The location of municipal sewage treatment plants, with accompanying wastewater marsh systems adjacent to urban centers, brings the possibility of potential major mosquito sources of both vector and pest species well within the mosquitoes' effective flight range.

Essentially the wastewater marsh system provides shallow

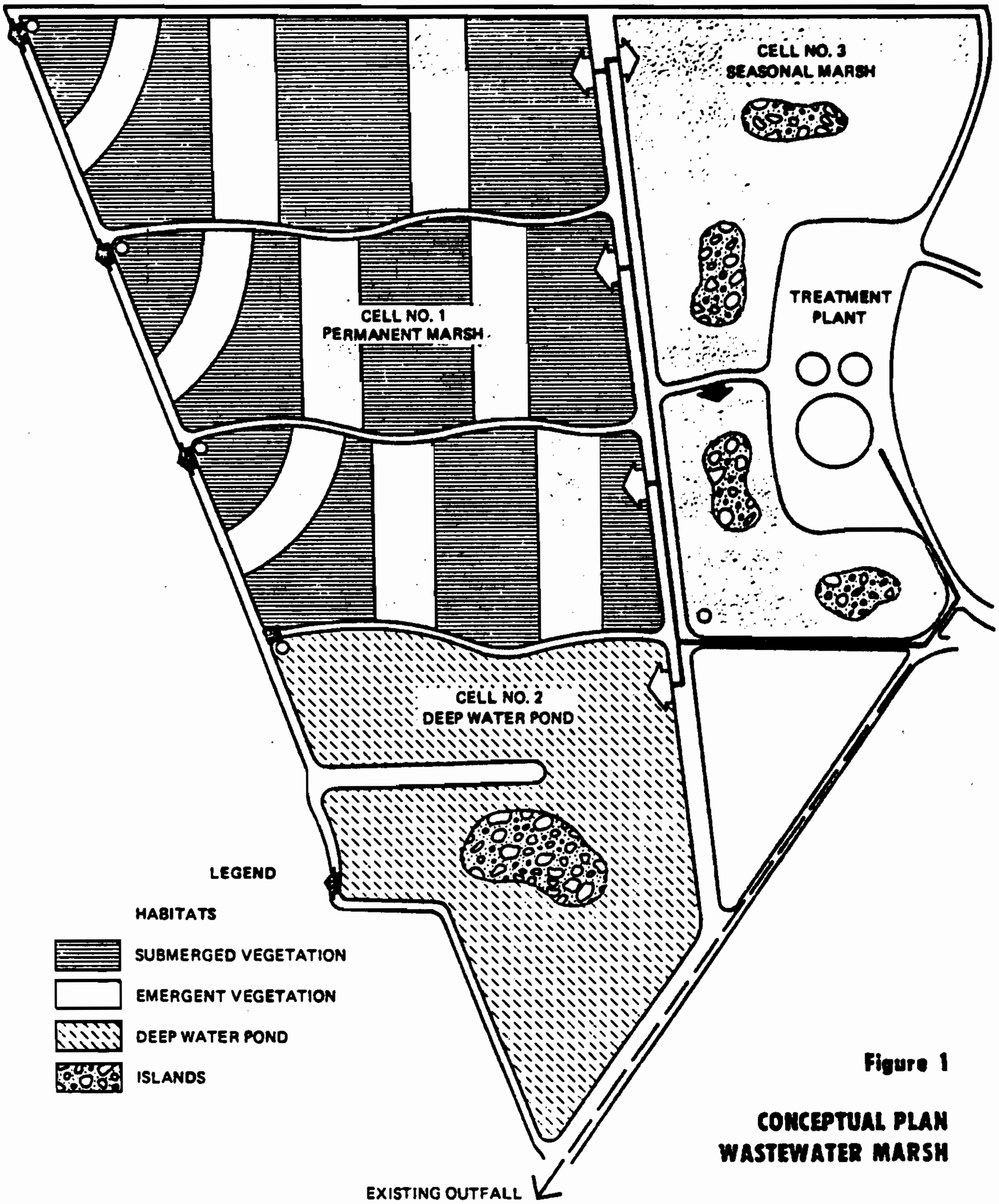

Figure 1

CONCEPTUAL PLAN WASTEWATER MARSH

water with various types of submerged or emerging vegetation habitats ideal for mosquito propagation. Studies of mosquito occurrence by Beadle and Harmston (1958), Smith and Enns (1967), of sewage stabilization ponds in the Midwestern states found maximum production of *Culex tarsalis* Coquillett and the *Culex pipiens* complex mosquitoes to occur in shallow ponds with a heavy density of vegetation, emergent/or submerged aquatic plants. The system's effluent can have high nutrient and organic levels that amplify the aquatic stage of the mosquito life cycle. Gerhardt (1959) studied the oviposition preferences of mosquitoes in shallow-water environments containing different levels of organic material. He concluded that the fermentation process at the soilwater interface caused the release of volatile organic acids that served as oviposition attractants for *Culex peus*, *Culex tarsalis* and *Anopheles freeborni*.

Basically, the wastewater marsh system is designed to operate on a year around basis which provides maximum opportunities for mosquitoes to infest the marsh.

Figure 1. shows a conceptual plan of a wastewater marsh system that is typical of what advocates of the system want to accomplish. There are three basic components in the system, deep water pond (Cell No. 2), permanent marsh (Cell No. 1) and seasonal marsh (Cell No. 3).

The deep water pond functions as an oxidation, storage and open water waterfowl impoundment. Mosquito problems would probably not occur in this kind of habitat if the inside levees were steep and free of heavy and overhanging vegetation. This pond would be an excellent method for culturing and maintaining mosquitofish, *Gambusia affinis*, that could be used in other parts of the system as an effective biological control agent.

The permanent marsh is made up of a series of small 5 to 10 acre plots that will be permanently inundated with 1 to 2 feet of wastewater. Submerged and emergent aquatic vegetation will be grown to provide the maximum vegetative water interface. This type of permanent marsh would have high probability of propagating mosquitoes.

The seasonal marsh, (Cell No. 3, Figure 1), is designed to be flooded with wastewater to a depth of about one foot in the early spring with repeated drying and reflooding to germinate and grow waterfowl food. The marsh could be dewatered in early summer, allowed to remain dry until reflooding in September or October to attract and feed migratory waterfowl. Mosquito problems of both the *Culex* and *Aedes* genera would most likely occur in this kind of marsh operation.

In 1981 the Vector Biology & Control Branch of California Department of Health Services, in cooperation with the Contra Costa Mosquito Abatement District and Mountain View Sanitary District, conducted a study of mosquito occurrence in a 20 acre wastewater marsh system that was developed by the Sanitary District in 1974. Bogard et al. 1982, (In press) indicated mosquito larvae occurred in densities of 8.5 to 11.0 larvae per dip in June from dense vegetation areas in Ponds 1A and B. These larvae were all identified as *Culex tarsalis*. Of the total number of 550 mosquito larvae collected by the dip method during the study period April-December, 1981, 82.5% were *Culex tarsalis*, 10% were *Culex erythrothorax*, the remainder were *Culex pipiens*, *Culex peus* and *Culiseta inornata*.

In more open areas of the marsh ponds where large numbers of mosquitofish occurred, mosquito larvae were difficult to find. Dense stands of cattails, tules and residual plant debris associated with this dense vegetation provided an ideal water-plant interface for harborage of large numbers of *Culex tarsalis* larvae protected from fish predators.

MOSQUITO CONTROL CONCERNS.–Mosquito control agencies need to play a major role in the initial planning and design of projects that include wastewater marsh systems as part of a community sewage treatment process. Mosquito prevention standards should be incorporated into the basic design and operation of a wastewater marsh system.

In 1978 the Vector Biology and Control Branch in cooperation with the California Mosquito and Vector Control Association, prepared criteria for mosquito prevention in wastewater reclamation projects. These criteria were prepared in response to changes in California water pollution regulations and the increased emphasis for reuse of wastewater. These reuse concepts for wastewater have serious prospects for mosquito production. The following criteria are used during the review of water quality discharge requirements, U.S. Army Corps of Engineers permits and environmental impact reports that relate to use of wastewater for wetland development. The criteria is based on ecological facts known to inhibit mosquito propagation.

Permanent Water Impoundments.

- Water depth should be a minimum of 4 feet.
- The inside slope of levees should be as steep as possible, preferably not less than 3 to 1.
- Ponds should not have small coves or irregularities around their perimeters.
- Top width of embankment should be a minimum of 12 feet to accomodate maintenance and inspection vechicles.
- Accumulations of debris, vegetation and algae mats should be routinely removed from the water surface.

Permanent and Seasonal Shallow Water Areas.

- Site preparation should include proper grading for rapid dewatering.
- Provide areas of deep water that can serve as mosquitofish holding ponds during the drawdown period.
- Provide proper water control structures, weirs, pumps and siphons for complete water management.

In shallow water areas the type and density of vegetation is critical in determining the effectiveness of mosquitofish. To maintain fish predation, the vegetative growth may have to be periodically removed or harvested. Another alternative for controlling vegetation would be varying the depth of water to discourage certain plant species.

The above criteria for mosquito prevention are general in nature. There is an important need for specific information on biological parameters, physical features and operational standards for developing wastewater marshes. A series of demonstration marshes are needed to study the biological and physical factors that would meet the needs of water quality, wildlife conservation and mosquito suppression objectives.

If the preventative approach is not closely adhered to, serious mosquito problems may develop in the community. If this occurs, the mosquito control agency may have to rely on pesticide control measures at a considerable cost to the municipality. Sections 2863 and 2807 of the California Health and Safety Code provide for a mechanism for the mosquito control agency to recapture control costs from the agency responsible for management of the wastewater marsh.

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A COMPARISON OF SAMPLING METHODS FOR ADULT *ANOPHELES FREEBORNI* AITKEN

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ABSTRACT

Five methods were compared for their effectiveness in sampling adult mosquitoes within orchards in Sutter County, California: (1) standard cubic foot red boxes, (2) larger "walk-in" red boxes, (3) CDC-CO_2 light traps, (4) a truck trap, and (5) animal baited traps. "Walk-in" red boxes were the most effective in collecting *An. freeborni*, while CDC-CO_2 traps collected the largest numbers of *Culex tarsalis* and *Aedes melanimon*. Standard red boxes placed at ground level collected greater numbers of *An. freeborni* than those placed at ca. one and two meters, while collections of *Cx. tarsalis* were similar for all three heights tested. "Walk-in" red boxes placed in 15 year old orchards collected more of both *An. freeborni* and *Cx. tarsalis* than boxes placed in seven year old orchards. A parity profile for *An. freeborni* and a summary of the metabolic states of the various mosquito species collected by the different sampling methods was compiled.

TOXICITY AND ATTRACTANCY OF THE HYDROPHYTE *MYRIOPHYLLUM* AGAINST MOSQUITOES

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During a study of chironomid midges in freshwater lakes in southern California, it was observed that the larval density of aquatic midges was inversely proportional to the density of the aquatic weed, Eurasian watermilfoil *(Myriophyllum spicatum)*. The Eurasian watermilfoil is a common nuisance species found throughout the United States. In testing the hypothesis that this weed was responsible for the midge reduction, Dhillon (1980) tested an extract of the weed against fourth instars of *Chironomus* and established an LC_{90} rated of 640 ppm.

Since extracts of *Myriophyllum spicatum* were effective against chironomids, we hypothesized that it might also be effective against immature mosquitoes. Other researchers have shown that aquatic plants can reduce mosquito populations (Angerilli 1980, Angerilli and Beirne 1974, Furlow and Hays 1972). This plant was collected from Spring Valley Lake in southern California where it is mechanically harvested as a nuisance weed. It was air dried in a greenhouse, chopped into small pieces, and stored in a freezer until needed. The plant was extracted with a benzene-methonal mixture, dissolved in acetone and then stored in a refrigerator until needed for testing.

Second- and fourth-instar mosquito larvae were exposed by placing 20 larvae in a cup with 100 ml water. Each cup of larvae was supplied with 0.05 g of ground up lab chow-yeast mixture. Forty-eight hours after the extract was applied, the percent mortality was recorded. Of the three species tested, we found that, for the second instar, the LC_{50} values were 8 ppm for *Aedes aegypti* L., 12 ppm for *Culex tarsalis* Coquillett, and 28 ppm for *Cx. quinquefasciatus* Say. The fourth instar was more tolerant to the material having an LC_{50} of 85 ppm for *Cx. tarsalis*, 96 ppm for *Cx. quinquefasciatus*, and 135 ppm for *Ae. aegypti*. At these concentrations, no delay in development was observed.

The pupae were tested similarly, with 20 in each cup. However, they were not given food, and mortality was observed after 24 hours. The pupal stage was the most susceptible stage in their development showing an LC_{50} of 5 ppm for *Cx. tarsalis* and 6 ppm in both *Cx. quinquefasciatus* and *Ae. aegypti*.

Experiments were conducted to better understand the nature of the plant extracts affecting mosquito pupae. In an experiment, three pans with identical surface area were each filled with 500, 100, and 1500 ml of water. One-hundred pupae were released in each pan and 150 µl of plant extract was added to each pan. After 24 hours, the percent mortality was nearly identical in all three pans irrespective of the concentrations of the plant extract. This shows that the controlling factor remains at the water surface. Other observations indicated that the pupae must come in contact with the surface film of this extract before mortality would occur.

During toxicity tests, a number of adult mosquitoes emerging from test and control cups were attracted to the cups containing *Myriophyllum* extract and drowned in them. Several experiments followed this interesting observation. Five-hundred pupae were placed in each of three spearate cages and allowed to emerge. Each cage contained two 100-ml cups of water and two 100-ml cups of water containing extract (250 ppm). In one cage, no food was added. The second cage contained a sugar-water soaked wick for mosquitoes to feed on. The third cage, which also contained suger water, was placed in a separate room containing vegetation to supply additional odors. Each day the mosquitoes trapped in the aqueous mixture of the extract were removed and counted.

With *Cx. quinquefasciatus*, 395 adults were collected from the plant extract in cages without food after four days. One-hundred seventeen adults were collected from the cage with sugar water, and 93 adults from the cage with sugar water and plant odors. Similar results are obtained with *Cx. tarsalis*. These results seem logical since the addition of a food source would reduce the number of mosquitoes going to the plant extract for food. Also, the addition of plant odors would help mask the odors coming from the plant extract. When *Ae. aegypti* was tested however, the results were surprisingly different. After four days, 145 adults were removed from the cage without food. From the cage with sugar water, 91 were removed. However, from the cage with sugar water and plant odors, 178 were removed. The same trend was observed in each of three replications. The addition of other plant odors apparently intensified the attractive factor present in the *Myriophyllum* extract. The nature of this attractancy will continue to be investigated.

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TECHNIQUES FOR A *GAMBUSIA AFFINIS* MARK-RECAPTURE STUDY

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ABSTRACT

Marking and detection techniques are presented for a *Gambusia affinis* mark-recapture study. The marking procedure described involves the subdermal injection of a fluorescent polystyrene pigment. Marking was relatively easy and the marks persisted well beyond the duration of the field study. Detection of marked individuals was simple and allowed for rapid counting and return of captured fish.

INTRODUCTION.–The mosquitofish (*Gambusia affinis*) has been widely used as a biological control agent for mosquitoes, however, several aspects of population biology are not known. Population levels have generally been characterized in relative terms. The purpose of our performing a mark-recapture study was to define, in quantitative terms, the population levels of mosquitofish in rice fields for the purpose of working toward an integrated pest management model.

In this paper a description of the marking procedures will be given as well as procedures for detection of marked individuals. Results concerning population levels are reported by Miura et al. (1982).

Marking techniques used for many fishes such as fin-clipping, tagging and tatooing are generally not appropriate for mosquitofish due to their small size. Methods previously used in mosquitofish marking studies are: Rhodamine B, O'Grady and Hoy (1972); Tetracycline drugs, DCAF, and polystyrene pigments, Vondracek et al. (1980). Based on the preliminary results of a mass marking study by Vondracek et al. (1980), we decided to use subdermal injection of a polystrene pigment.

MATERIALS AND METHODS.–The fish used for marking were a mixture of field and sewer farm reared fish. We wanted to use fish from the field in which we were to release the marked individuals but not enough were available.

For marking we used fish of moderate size with those over 40 mm (SL) perferred. Both sexes were used but females accounted for the largest part of the marking stocks. The fish were held in the laboratory one week prior to marking to acclimate to holding conditions and to observe mortality. Acclimation and holding conditions were 22 ± 1°C and 16 h light - 8 h dark photoperiod.

The dye used was a fine powdered fluorescent polystyrene pigment in a melamine-sulfonamide-formaldehyde resin. When the pigment is exposed to UV light it fluoresces but small quantities in normal indoor light and sunlight have no apparent color. The dye was purchased from Scientific Marking Materials in Seattle, Washington.

The general marking technique involved high pressure subdermal injection of the dye. In order to force the dye into the fish tissues we used a sandblast gun with hopper reservoir. The sandblast gun and hopper were made by Speedaire.®

The gun was attached to a compressed air source to provide the pressure for subdermal dye injection. We used a large air compressor with a twenty five gallon storage tank. A high pressure setting was necessary to achieve good marking results.

The fish to be marked were placed inside a wooden framed screen box. Screen on the bottom of the box was 1/16" mesh plastic window screen and the top was 1/4" mesh hardware cloth. The screen allowed passage of excess dye past the fish and kept them confined during application.

Spraying time was ten seconds with the sandblast gun ten to twelve inches from the fish. Tank pressure at the start of marking was 175 psi and dropped to about 160 psi. The actual pressure at the gun tip was significantly less than the tank gauge indicated but was not measured.

After application of the dye, the fish were immediately rinsed off with water to remove excess material. Ten seconds of dye application completely coated the fish and they appeared solid colored. Following rinsing the fish were placed in a seventy five gallon holding tank with treated water. The water was treated with 2 ppm potassium permanganate and 500 ppm sodium chloride to prevent infection. Water in the holding tank was aerated and filtered through a biological filter and changed daily. Retreatment with sodium chloride was made daily. Dead fish were removed when found. Holding time after dye application was four days prior to field release. Healthy fish were counted and released into a rice field for recapture at a later time.

One of the objectives of our study was to be able to determine marked from unmarked fish in the field and to return them in as little time as possible. We constructed a viewing box with field portability for this purpose.

The viewing box was built around a four liter glass aquarium measuring 6" x 10" x 7" into which the fish were placed for mark detection. Aluminum foil was used to cover the out-

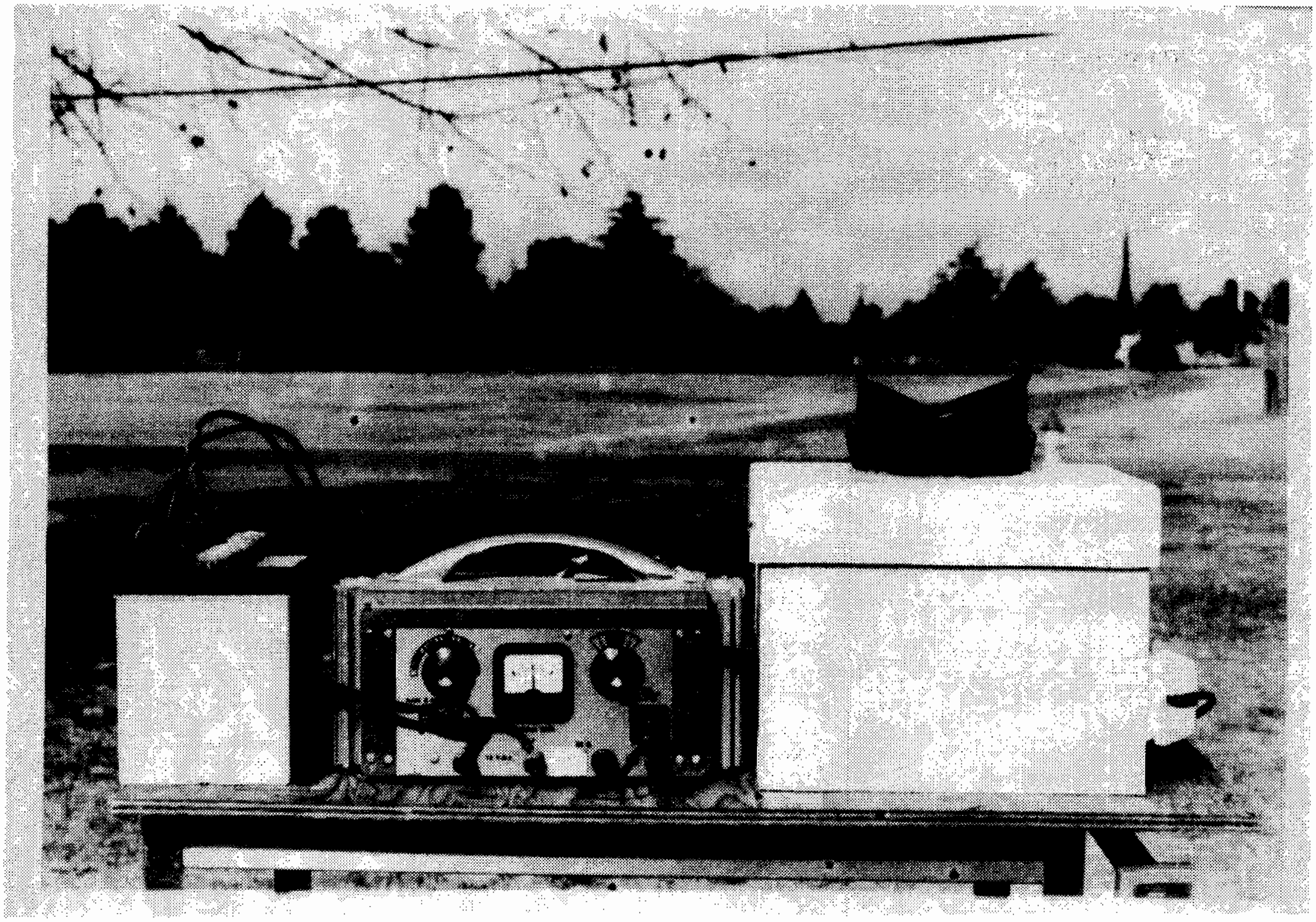

Figure 1. Field apparatus for detection of marked fish. Far right, viewing box with UV light source. Middle, inverter to convert DC battery current to AC for the light source.

side of three sides of the aquarium in order to brighten the lighted area and make it easier to pick out marked fish. At a later time we replaced the foil with mirrors, which worked better. The light source was a long wave black light (Black Ray UVL-22 from Ultra-violet Products). Figure 1 shows the viewing box and power source.

To power the light source in the field we used a twelve volt battery and an inverter to provide the AC current necessary. The small battery we used was able to provide power for more than five hours.

The procedure followed in the field was to remove the fish from the traps and place them in buckets. Up to about twenty fish netted out of the buckets could be placed in the viewing aquarium at one time. Marked fish were easy to pick out from unmarked individuals. As soon as one group was counted it was returned to a bucket and when counting was complete for fish from one group of traps they were returned to the capture area. It usually took less than three hours to collect and count the fish from twenty five traps.

RESULTS AND DISSCUSSION.–During the premarking acclimation period there was less than 5% mortality to the marking stocks. Most mortality was noticed in the smaller fish and males. The post marking mortality was 10-20% depending on the size of fish used in the particular marking batch. Larger fish survived better than small ones and males did not survive as well as females. Most mortality occurred in the twenty four hour period after marking with a estimated 1-2% thereafter. With an initial marking stock of 1100 fish over 900 survived.

Marking success was determined to be good with over 90% of the surviving fish having clearly visable marks. In order to check for mark retention, twenty five marked fish which were recaptured were brought back to the laboratory for observation. These fish which were marked July 13, 1981 had clearly visable marks still present on April 20, 1982.

The techniques used allowed for a rapid determination of marked from unmarked fish in the field. Marking was relatively easy and required no complicated apparatus. Fish mortality due to marking was not excessive and mark retention was determined to be excellent.

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CULEX TARSALIS COQUILLETT: TEMPORAL CHANGES IN FEMALE RELATIVE ABUNDANCE, REPRODUCTIVE STATUS AND SURVIVORSHIP

William K. Reisen and Marilyn M. Milby

ABSTRACT

Temporal variations in the relative abundance and reproductive status of a female *Culex tarsalis* population were studied in the arid Sierra-Nevada foothills of Kern County, California during the spring, summer and fall of 1981. Relative abundance patterns in light traps and in red box shelters were positively correlated throughout and covaried positively with temperature and *Cx. tarsalis* abundance in light traps operated at 22 other sites in rural Kern County. The proportions of empty, virgin and parous females remained relatively constant from May through late August, after which the proportions of empty and virgin females increased markedly. During autumn the population bifurcated into reproductively inactive and active components. Release-recapture and laboratory observations indicated that the duration of the nulliparous period during the summer was six days and the gonotrophic cycle lasted four days. Survivorship of resting females during May-August was estimated to be 0.74 and 0.87 using two calculation methods. These estimates were related to the potential for arbovirus transmission.

AGE CHARACTERISTICS OF MIGRATING *AEDES DORSALIS* IN SALT LAKE CITY, UTAH, SEPTEMBER 1981

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Last summer migrations of *Aedes dorsalis* from salt marshes and duck clubs north and west of Salt Lake City are known to occur but few actual reports have been published. Rees (Univ. Utah Bull. 25(5): 1-6. 1935) gave a full account of the daily progress of a mass movement of *Ae. dorsalis* that took place 50 years ago. An unusually heavy rainfall at the end of August resulted in an enormous brood of mosquitoes. The flight of adults was traced from the emergence area through points in Salt Lake City and up one canyon to 22 miles southeast of the source 10 days after emergence.

From 16 September to 2 October 1981, a dispersal of *Ae. dorsalis* was observed over a period of 17 days. Females collected in and around Salt Lake City were dissected to gain information about the physiological age of migrating *Ae. dorsalis*.

Localized storms within the Salt Lake and Utah Valleys can produce great amounts of water that are funneled by the Jordan River, canals, and other drainage systems onto the marshes at the southeast end of the Great Salt Lake. Flooding of some of the marshlands, including duck clubs, began the last half of August when temperatures were as much as 13°F above normal. Extensive acreages were flooded to produce hordes of *Ae. dorsalis*.

Complaints were received from residents in the northwest part of the City beginning 1 September. From 11 to 16 September complaints came from the northwest and east section of the City.

Four New Jersey-type light traps within reasonable range of the emergence area attracted *Ae. dorsalis* throughout the month of September. Starting on 14 September and for the next few nights, there was an increase in the number of *Ae. dorsalis* females caught. Except for one trap, male *Ae. dorsalis* were present at that time and on some nights until the end of the month. Three traps closer to the City had a smaller increase in the number of female *Ae. dorsalis* at that time but there were no males. Concurrently, three traps across the City to the east and south collected small numbers of *Ae. dorsalis* females. One trap, about 12 miles from the emergence area, continued to lure a few *Ae. dorsalis* females through 20 September.

Aspirator collections of live, adult females were initiated on 16 September. Biting *Ae. dorsalis* were already in the City. There were 30 collections, made irregularly at varied locations, spanning 17 days. Collecting places were combined into zones according to distances from the breeding grounds:

	Distance	Number of Locations	Number of Collections	Number Dissections
Zone 1 - close	0-4 miles	8	11	292
Zone 2 - intermediate	4-6 miles	5	10	194
Zone 3- distant	6-12 miles	5	9	86

Five hundred and seventy two total female *Ae. dorsalis* were dissected. All were post-teneral.

After numerous dissections showing every female to be inseminated, further examinations for presence of sperm were discontinued except for unusual specimens such as those that were parous or parasitized. It was assumed that mating had taken place at the emergence site.

Evidence of nectar feeding within the previous 12 hours was found in 96% of the females tested for fructose by the anthrone reaction (van Handel, Mosquito News 32: 458. 1972). The samples were from within the City and from intermediate northwest locations.

The majority of the females had not developed their first batch of eggs. Most had ovaries resting in stages I or II with an empty midgut. They were evidently seeking a blood meal. These nulliparous females were found in close and intermediate zones for 17 days thus indicating continued emergence from different places. It would not be expected that blood feeding and egg maturation be delayed more than a few days after becoming post-teneral. Nine females with fresh blood had, no doubt, taken it from the collector. Seven females with advanced degrees of digested blood would soon have deposited eggs.

There was no evidence of oviposition having taken place until after 20 September. Of the total number dissected, about 20% were parous. These tended to be distributed in relation to distance from the source. Ones furthest from the flooded marshes were found in much smaller collectible numbers with only 7% older than the first ovarian cycle. Five had recently oviposited as shown by stretched pedicels on the ovarioles. As unlikely as it may seem that *Ae. dorsalis* eggs would survive in a cemetery or park habitat, on two occasions in past years, *Ae. dorsalis* larvae and pupae were taken during routine inspections in June prior to putting fish in ornamental pools on the north bench of Salt Lake City. No mosquitoes were taken in the City after 27 September.

In the intermediate zone, 10% of the females were parous.

At sites closer to the emergence areas, 30% had laid eggs including one female on 1 October and another on 2 October in their third ovarian cycles, having accomplished two ovipositions. Judging by these collections, greater physiological longevity and reproductive potential are achieved by mosquitoes that remain close to the breeding area.

All parous females were inseminated.

On 1 October, three unusual females were found in a collection from a site close to the marshes. They were post-teneral, inseminated, and with empty midguts. The oviducts were not stretched. Instead of ovarioles, the ovaries were packed with what was presumed to be sporangia of the parasitic fungus, *Coelomomyces*.

During the migration of *Ae. dorsalis*, males were reportedly seen by one of the authors but none was collected.

The implications are that when tremendous broods of mosquitoes emerge, some are stimulated to leave the area even though blood sources are easily available and there are ample oviposition sites. Sufficient numbers of mosquitoes remain at the breeding site to replenish the stock.

There is no firm evidence but it appears that if *Ae. dorsalis* are ready to fly distances, they will do so under atmospheric conditions of higher-than-normal percent relative humidity, a slight prevailing breeze, with warm, bright moonlit nights.

The flight path of the mosquitoes from the salt marshes through Salt Lake City to a point 12 miles southeast of the nearest source was exactly comparable to Rees' observations made in 1932. Changes in the environment caused by man's activities, i.e. growth of the City, have not altered the dispersal pattern of *Ae. dorsalis*.

INTERCANYON MOVEMENT OF MARKED *CULEX TARSALIS*

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ABSTRACT

Approximately 3,000 one - two day old *Culex tarsalis* of each sex, reared from field-collected pupae, were marked with fluorescent dust and released in each of three adjacent Sierra Nevada foothill canyons in August 1981 to study population exchange. Recapture collections were made for ten consecutive days following the first release and utilized three methods: 14 CO_2-augmented CDC traps with lights and four without lights, 12 small and five walkin red box shelters, and aerial net sweeping of male swarms. Most recaptured females from the first two days' releases were dissected to determine their reproductive status. Temperatures during the recovery period averaged 32°C (range = 21°C to 44°C).

Males failed to disperse from the canyon in which they were released. This had been noted in previous studies at the same site which involved the release of radiosterilized males from both field-derived and laboratory populations.

Females moved freely among the canyons; marked immigrants were more likely than non-migrants to have mated prior to recapture. All dissected marked females were mated by the fourth day following release. The median time from emergence to first oviposition was five days. Parity rates were generally lower for females collected in shelters than for those in trap collections. Autogeny in the native population was 44% at this time and may have resulted in undersampling of nullipars in traps.

Daily loss rates were estimated horizontally by the regression method for marked adults and vertically from the proportion parous for unmarked females. Loss rates were higher for marked males (22-38%) than for marked females (24-31%). The loss rate for unmarked females in the central canyon was quite low (14%); this canyon received more immigrants than either peripheral canyon and thus seemed to support an older population.

The failure of males to disperse from their release sites coupled with the observation that females tend to mate prior to emigration demonstrates the importance of choosing multiple and dispersed sites for release in genetic control trials. Future studies using genetically altered males at this site will have to utilize releases in all three canyons to compensate for intercanyon movement. This should increase the degree of contact between released males and virgins from the target population.

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EFFECTS OF IMMATURE REARING CONDITIONS ON THE SIZE OF ADULT *CULEX TARSALIS*[1]

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ABSTRACT

Recent studies under laboratory and field conditions have shown that adult mosquito size is affected by several factors during larval growth stages. In order to evaluate the impact of three of these factors, a laboratory colony of *Culex tarsalis* was reared under three temperatures (18°, 26°, and 31°C), three densities (600, 1200, and 2400 larvae per pan), and three variations of a basic food regime (.3BR, BR, and 3BR). Size at death was determined for cohorts of 25 males and 25 females from each replicate of each rearing condition.

Temperature, food, and density significantly influenced adult wing length as well as development time from first instar to adult. The highest temperature, which produced the smallest mosquitoes, also had the fastest development time. The medium and low temperatures produced progressively larger mosquitoes and had progressively longer development times. However, the opposite was true for the food treatments, where the 3BR group produced the largest mosquitoes but had the fastest development time. Density did not show a clear trend, other than the fact that the 600/pan group were consistently larger and took fewer days to develop.

Survival and development time decreased as a function of increasing water temperature. Developmental time decreased and survival increased as the amount of food per larva increased in the food and density experiments.

The autogeny rate was influenced solely by food, the 3BR group being the highest at 33%. Longevity also was significantly related to size. Throughout all of the treatments, larger adults tended to live longer than small adults.

Our data would seem to support the concept of a developmental "window" through which larvae must pass in order to successfully pupate. This window appears to result from the interaction of the amount of food ingested per unit of developmental time.

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A PRELIMINARY SIMULATION OF MOSQUITO CONTROL IN COYOTE HILLS FRESHWATER MARSH, ALAMEDA COUNTY, CALIFORNIA

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Three often-cited reasons for attempting to model ecosystems are (a) to aid in the conceptualization of the system, (b) for the prediction and/or management of the system, and (c) to formalize present knowledge of the system. This project contemplates more effective management of a freshwater marsh and is currently in the conceptualization stage of a predator-prey model for such a marsh.

The modelling of aquatic ecosystems is in its infancy. Complex aquatic models have been developed for salt marshes (Hopkins and Day, 1977; Pomeroy and Wiegart, 1981) and freshwater reservoirs (Patten, 1976). This paper deals with the first phase of the modelling program for the freshwater marsh. Our model begins by examining the crucial relationship between the mosquitofish, *Gambusia affinis*, and one of its prey items, larvae of the mosquito *Anopheles freeborni*. The relationship will form the nucleus of a more complete marsh model.

Modelling the mosquitofish as a predator on mosquito larvae has recently come under intensive study in California habitats, especially rice fields (Cech et al. 1980 a,b; Cech et al. 1981). These models have been primarily bioenergetic models of the mosquitofish examining aspects of prey-selection, metabolic rate, growth rate and population dynamics. These bioenergetic models were also stimulated by the desire to control or manage mosquito populations using *Gambusia*, especially in the rice fields of central California. As stated earlier, our project is motivated by a similar desire, only the model being developed here approaches the predator-prey problem from a different perspective. The future synthesis of the two approaches is anxiously awaited. Our model presently outlines a few of the important factors influencing the effectiveness of mosquitofish in controlling mosquitoes and how those factors could be used by vector control managers to simulate predator-prey populations.

The Coyote Hills Freshwater Marsh is located in Fremont, California adjacent to South San Francisco Bay. In the month of May the marsh is approximately 14-15 acres in size. During the peak winter rain runoff period, the area is used as a ponding area by the Alameda County Flood Control District. After the rains have subsided, management of the marsh becomes the responsibility of the East Bay Regional Park District. The park district uses the marsh as an educational site, mainly for bird watching and environmental observations. Their main wildlife concerns deal with marsh dependent birds and migratory waterfowl.

In the month of May the marsh is approximately 14-15 acres in size. By July the size of the marsh usually decreases to approximately 4 acres. Water depth varies from nearly 4 feet in April to 4-6 inches from July through September or October. Rainwater is the source of freshwater during the winter and spring months. During the summer, usually by July, the marsh would characteristically dry up. The park district purchases water and pumps it into the marsh system to maintain the marsh through the dry period. In some years, however, there has been significant fish kills before adequate water has been supplied.

Vegetation of the marsh includes cattails, (*Typha*), and two submergents, the sago pondweed (*Potamogeton pectinatus*) and traces of *Myriophyllum* spp. The first two plants are characteristic of mosquito breeding habitats in the area. The Coyote Hills Marsh is capable of producing high levels of mosquitoes each season. The species produced include *Anopheles freeborni, An. punctipennis, Culex tarsalis, Cx. erythrothorax, Culiseta incidens* and *Cu. inornata*.

The model is divided into one abiotic and five biotic components (Table 1.). Carbon flows are designated by solid lines while information flows are shown as dashed lines. The model is designed to simulate the predation of mosquitofish on mosquito larvae and alternative prey in the freshwater marsh. The impact of the mosquitofish on the mosquito larvae is indirectly influenced by pond depth in two ways. The first is via predation by wading birds. As pond depth increases fewer mosquitofish are exposed to predation by birds. The second influence of pond depth is its influence on the growth of pondweed. Anderson (1978) in plotting *Potamogeton* production versus depth found a steep bell shaped curve with maximum biomass at approximately 60 cm in depth with a sharp decline in deeper or shallower waters. As pondweed biomass increases portions of the marsh surface become occluded and the rates of encounter between predators and prey are assumed to decrease. Therefore average marsh depth will influence the effectiveness of mosquitofish in controlling the mosquito population, with the least control possible at depths of approximately 60 cm. Specific values assumed for growth rates, searching rates, assimilation efficiencies, predation rates, and carrying capacity, can be found in the appendix.

There are a few interactive simulation options built into the modelling program. Initial values of mosquitofish density, mosquito larval density and average pond depth are specified prior to each simulation. Management options which may be exercised prior to each simulation are the stocking of mosquitofish and/or the application of pesticides. Stocking of mosquitofish can be done to any level, but presently is

Table 1. Mosquito larvae and mosquitofish densities (a) and pondweed phenology (b) during simulations at three different marsh depths.

Table 1a

Date	Mosquito larvae ($mg \cdot m^{-2}$)			Mosquitofish ($mg \cdot m^{-2}$)		
	15cm	50cm	85cm	15cm	50cm	85cm
May 15	0.20	0.20	0.20	33	33	33
June 14	0.01	0.01	0.01	33	45	45
July 14	0.02	0.01	0.01	6	52	52
August 13	0.21	0.06	0.01	4	59	59
September 12	1.90	1.06	0.01	6	67	67
October 12	10.96	17.52	0.01	8	77	76

Table 1b

Category	Depth		
	15cm	50cm	85cm
Pondweed reaches water surface	June 15	July 1	July 15
Pondweed reaches peak standing crop	July 15	July 31	August 14
Pondweed yield ($g \cdot m^{-2}$)	10.4	169.5	66.4

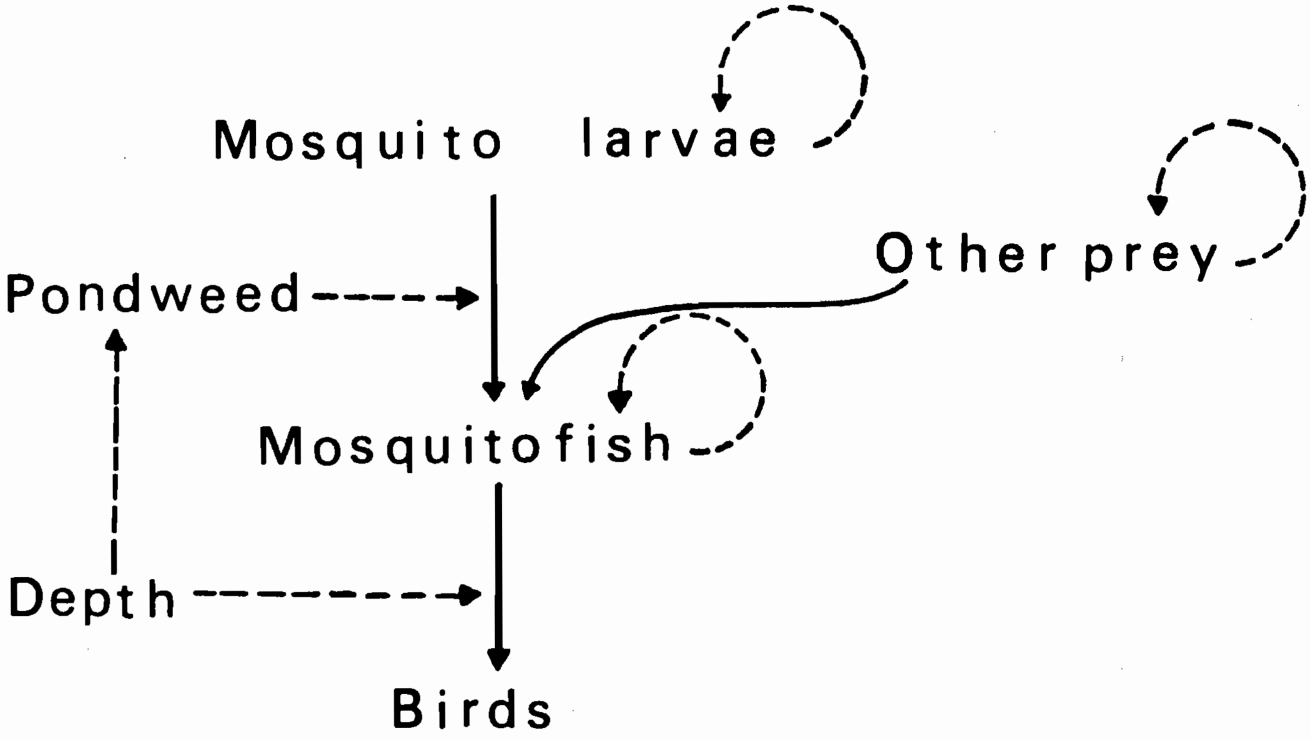

Figure 1. The basic predator-prey model with carbon flows shown as solid lines and information flows as dashed lines.

only done on day 17 of the 155 day simulation. Pesticide applications may be triggered in the simulation automatically when the mosquito larvae exceed the critical density (threshold) specified before each simulation. Both fish stocking and pesticide applications include cost functions which are totalled at the end of each simulation. Stocking costs are based on the numbers of truck loads of fish required to reach the specified stocking level. Pesticide cost is computed at a fixed cost per application. The total biomass of pondweed produced during the simulation is also given because of its possible significance in waterfowl management in the system.

The current model is written in basic and designed to easily operate on most personal microcomputers. This is considered an important aspect of any potential management tool in keeping it accessible to field users. The results presented here were run on a PDP11 with RY8T Operating System which is part of the computing facilities at California State University, Hayward.

RESULTS.–A series of simulations were run in order to demonstrate the model features of depth/pondweed effects, fish stocking and pesticide application.

The first simulations were a depth series. The influence of pond depth on predator-prey dynamics, as discussed earlier, is via pondweed growth which reduces encounter rates. The three depths used were 15, 50 and 85 cm all beginning with the same density of fish (0.3 lbs/acre) and mosquito larvae (0.2 mg/meter sqd.). The results are shown in Table 2. Note the differences in the time the pondweed reaches the surface (P W Surface) and the peak standing crop (Peak S C) for the three depths. In the 85 cm simulation the lack of any significant reduction of fish-mosquito encounter rate keeps the mosquito population at the lower boundary of the model where the alternative invertebrate prey are maintaining the fish population. Maximum occlusion occurs at the 50 cm depth where the pondweed reaches its peak density. The relationship between the mosquito levels and pond depth appears consistent with field populations from Alameda County.

The second simulation series was a fish stocking series done at a constant marsh depth of 50 cm (Table 3). The purpose of this series of simulations was to demonstrate the management decision option of fish stocking and marsh depth which was shown, in the first series of simulations, to produce a high population of mosquitoes. The results, Table 3, indicate that the mosquito population reaches high levels at the end of the season even at very high stocking rates. However, control early in the season, prior to pondweed occlusion, fish could be extremely effective.

The third simulation series was designed to examine the control and total costs of a combined fish stocking and pesticide spraying management plan. The permutations are staggering if the managers can control depth, stocking rate and larval threshold levels. These levels were done at only one depth 50 cm for three stocking densities and two critical mosquito densities (Tables 4 and 5). Because of the simple cost functions of the model it is clear that under all circumstances spraying is less expensive than stocking fish for controlling mosquitoes. The costs of each used here are only

Table 2. Mosquito larvae and mosquitofish densities during simulations done at 50cm marsh depth with three fish stocking rates. All stocking was done on 14 June to the levels shown.

Date	Mosquito larvae ($mg{\cdot}m^{-2}$)			Mosquitofish ($mg{\cdot}m^{-2}$)		
	Run 1	Run 2	Run 3	Run 1	Run 2	Run 3
May 15	0.20	0.20	0.02	33	33	33
June 1	0.01	0.01	0.01	39	89	177
June 14	0.01	0.01	0.01	45	96	188
July 14	0.01	0.01	0.01	52	109	215
August 13	0.06	0.03	0.02	59	124	243
September 12	1.06	0.41	0.10	67	141	277
October 12	17.52	4.03	0.33	77	160	312

Table 3. Mosquito larvae densities during six simulations with variable management plans (2 pesticide spraying levies X 3 fish stocking rates) at the same depth) 50cm. Pesticide spraying thresholds ($mg{\cdot}m^{-2}$) L = 0.5, H = 5.0; fish stocking rates ($mg{\cdot}m^{-2}$) N = 0, M = 89, H = 177.

Date	Mosquito Biomass ($mg{\cdot}m^{-2}$)					
	LN	LM	LH	HN	HM	HH
May 15	0.20	0.20	0.20	0.20	0.20	0.20
June 14	0.01	0.01	0.01	0.01	0.01	0.01
July 14	0.01	0.01	0.01	0.01	0.01	0.01
August 13	0.06 *	0.04	0.02	0.06	0.02	0.02
September 12	0.02	0.41 *	0.10	1.06 *	0.41	0.10
October 12	0.32 *	0.07	0.33	0.19	4.03	0.33

*Spraying during this interval

Table 4. Cost breakdown for the six management plans shown in Table 3.

Item	Cost (dollars)					
	LN	LM	LH	HN	HM	HH
Stocking	0	1,680	4,200	0	1,680	4,200
Spraying	630	315	0	315	0	0
Total	630	1,995	4,200	315	1,680	4,200

Table 5. Appendix. Modelling equations and assumptions.

```
240 REM **** MODEL DESCRIPTION, ASSUMPTIONS, VALUES, ETC. ****
250 !
260 REM ****            ANOPHELES FREEBORNI                ****
265! !
270 !       DN1/DT = THE NET INCREASE IN POPULATION BIOMASS
280 !       Q1 = BIOMASS OF LARVAL POPULATION (Q1 = Q1+DN1/DT)
290 !       DN1/DT = R N1 (1-N1/K)     WHERE :
295 !       DN1/DT = RATE OF CHANGE OF LARV. POPULATION (1 DAY)
300 !       R1 = NET GROWTH RATE PER INDIVIDUAL
310 !       N1 = NUMBER OF INDIVIDUALS
320 !       K = CARRYING CAPACITY, MAX THAT ENVIRON SUPPORTS
330 !
340 !
350 REM *****  ASSUMPTIONS ON ANOPHELES FREEBORNI          *****
360 !
370 !        (1). R = .12 /DAY
380 !        (2). N1 INITIALLY DETERMINED BY STANDARD DIPPING
390 !             ONE DIP = SAMPLE OF 1/10 OF A METER SQUARED
400 !             ASSUME EACH LARVAE WEIGHS .1 MG.
410 !             THUS IF 20 LARVAE/DIP THEN INITIAL
420 !             BIOMASS = ..0001 G/M (200 LARVAE/M) = .02 G/M
430 !        (3). K = .2  (ASSUME 200 LARVAE/DIP = K)
440 !             K = .2 G/M
450 !
460 REM ****            OTHER PREY EATEN BY GAMBUSIA     ****
470 !
475 !      R3 = .12 GROWTH RATE OF OTHER PREY
480 !
490 REM ****                 GAMBUSIA AFFINIS            ****
500 !
510 !       DN2/DT = (P N2)(A1) + (N3)(A2) - D       WHERE:
520 !       DN2/DT = NET INCREASE IN POPULATION BIOMASS (1 DAY)
530 !       N2 = N2 + DN2/DT (USE TO SOLVE NUMERICALLY)
540 !       P = A Q N1       WHERE :
```

Table 5. continued.

```
550 !        A = % OF LARVAE EATEN /# ENCOUNTEERED
560 !        Q = SEARCHING RATE - AREA SEARCHED IN UNIT TIME
570 !        N1 = DENSITY OF LARVAE - NUMBER/METER SQUARED
580 !        N2 = DENSITY OF GAMBUSIA - #/METER SQUARED
590 !        A1 = ASSIMILATION RATE WHEN PREY IS LARVAE
600 !        A2 = ASSIMILATION RATE WHEN PREY IS OTHER
610 !        N3 = DENSITY OF OTHER PREY
620 !        QD = ADJUSTED RATE OF PREDATION DUE TO PONDWEED
630 !
640 REM ****        ASSUMPTIONS - GAMBUSIA AFFINIS        ****
645 !
650 !        (1). A = .05 ASSUMED % LARVAE EATEN PER ENCOUNTER
660 !        (2). Q = 100 ASSUMES 100 SQUARE METERS SEARCHED IN DAY
670 !        (3). A1 = .12 ASSUMES 12% ASSIMILATION EFFICIENCY
680 !        (4). A2= .10 ASSUMES 10% ASSIMILATION EFFICIENCY
690 !        (5). D = .008 PER DAY RATE OF DEATH (PREDATION ETC.)
700 !        (6). AVE. GAMBUSIA WEIGHS .567 GRAMS
710 !        (7). QD= ((-.572082 * Y1) + 100)/100      WHERE:
720 !             QD = ADJUSTED PREDATION RATE
730 !             Y1 = BIOMASS OF PONDWEED
740 !        (8). K2 = 8.30809   CARRYING CAPACITY 75 LBS/AC.
750 !
760 REM ****        PONDWEED = POTAMOGETON PECTINATUS      ****
765 !
770 !              FROM ANDERSON , ECOLOGY, VOL.59, NO.1
775 !
780 !        Y1 = (11.5 * X1) - (.107 * X1 ** 2) - 137.98      WHERE :
790 !        Y1 = BIOMASS OF PONDWEED G/M , WHERE 0<= Y1 <= 174.8
800 !        X1 = WATER DEPTH , WHERE 13.76 <= X1 <= 93.71
810 !        DA = 31 + QS  WHERE: IN 31 DAYS FROM MAY 1ST THE
820 !             PONDWEED BEGINS TO GROW AND IN QS DAYS IT
830 !             REACHES THE SURFACE.
840 !        QS = (.430657 * X1) - 5.9
845 !
850 REM ****        ASSUMPTIONS - PONDWEED                ****
855 !
860 !        1. ASSUME GROWTH BEGINS ON MAY 15TH (FROM TUBERS)
870 !        2. ASSUME PLANTS REACH WATER SURFACE JUNE 15TH TO JULY 1ST
880 !        3. ASSUME PEAK STANDING CROP BY JULY 15TH TO AUGUST 1ST
890 !        4. ASSUME PLANTS DIE BACK OCTOBER 15TH
900 !        5. ASSUME PLANTS DISAPPEAR BY JANUARY 1ST
910 !        6. ASSUME PLANTS IN SHALLOW WATER REACH SURFACE SOONER
920 !        7. ASSUME QS = (.430657 * X1) - 5.9 PREDICTS THE NUMBER OF DAYS
930 !           REQUIRED TO REACH WATER SURFACE
940 !        8. ASSUME MAXIMUM INTERSECTION OF SURFACE OCCURS
950 !           IN 30 DAYS.
960 !
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those immediately paid by the marsh manager. Costing, function can easily be manipulated to included regional and seasonal differences in the costs of both activities. Field experimentation would allow the calculation of residual benefits during the next season from stocked fish, however the residual costs to the ecosystem of pesticide application is considered a much more difficult calculation.

The final result we would like to report is the informal feedback we have recieved from our colleagues on the utility of the model to management. During a workshop on "Systems Modelling for Vector Control" twenty vector control professionals experimented with this demonstration model. The workshop was sponsored by the Northern California Chapter of the Society of Vector Ecologists and was held on campus of California State University, Hayward on 20 November 1981. The feedback we were most concerned with were comments on ease of operation, logic, and ease of the interpretation of the simulation output. Comments on all three areas were all very positive with several users suggesting we include graphical output in addition to the present tabular output. Most users felt the model could be an important management tool provided the validity of the model is clearly established using field data and experimentation.

FUTURE DEVELOPMENTS.–The first modifications of the model will include the effects of temperature on all transfers; mosquito growth rates; prey selection behavior in mosquitofish and size variations in the mosquitofish population. Much of the information has been worked out by Cech and co-workers at U.C. Davis (Cech et al. 1980 a, b; Cech et al. 1981). However, the application of some of the published literature to freshwater marsh populations needs to be tested experimentally. Some of the experiments planned include studies of prey selection by mosquitofish and providing details on the indirect interactions between mosquitofish and the mosquito larvae. Murdock (1981) has found evidence for indirect interactions which might be the result of mosquitofish selecting prey items normally preferred by invertebrate predators thereby increasing the subsequent numbers of mosquito larvae eaten by invertebrate predators. This could be an extremely important process if considering that we may be establishing residual trophic control over the mosquito population by having high initial fish densities. This control would be important if heavy vegetational cover later reduced encounters between fish and mosquito larvae. Other research projects planned include the phenology of pondweed in the Coyote Hills Marsh and in perhaps a few years experimentation on water level manipulations in freshwater marshes being created in Hayward. Both of these will be important modelling contributions because of the high sensitivity of the model to depth and pondweed variations.

Some of the most obvious interactive and management options not yet in the model include varying the day of fish stocking and to generate a variable cost function for spraying. The later would be appropriate where the costs varied during the season because of say overtime during the busy season or shortage in equipment or materials. (As an example it might cost twice as much to spray in July as it does in May.)

In summary, the model appears to reasonably reflect the observed relationship between predator and prey and the way this relationship is altered by change in marsh morphology. The interactive options gave resource managers familiar with the natural history and day-to-day management of similar systems a clear representation of the relative impact of a variety of realistic management options.

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THE IMPLICATIONS OF A FRESHWATER SIMULATION MODEL ON THE ALAMEDA COUNTY MOSQUITO ABATEMENT DISTRICT

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The use of computers has already had a profound impact on the programs of the Alameda County Mosquito Abatement District. Computers now store and process operational data in the District, and generate timely reports for decision makers. It is now generally accepted that the automated system processes more data, more quickly and accurately than was accomplished by the previous manual system.

As presented in the previous paper, the District is in the early stages of developing a simulation model of a freshwater system. The computer in this case is doing more than simply providing information to decision makers, the Coyote Hills Model is designed to predict the consequences of alternative control approaches. Here the computer is encroaching into the decision-making-process, a revolutionary change in the District's approach to mosquito control. There is no doubt in my mind that we are entering a new and exiting era in vector control with both negative and positive implications.

POSITIVE IMPLICATIONS.–The ultimate objective of the Coyote Hills Model is to increase the effectiveness and efficiency of mosquito control. The previous presentation by Dr. Schooley has shown you the way the model operates. If the model can be validated, it will assist park district personnel and our district personnel is finding a management scheme that would maximize wildlife and recreational benefits while adequately controlling mosquito populations. The model provides the opportunity to try out various management strategies and to select an optimum approach. In a very short period of time, the model enables us to test various water depths, fish stocking rates and thresholds for pesticide applications, all by simulation rather than by costly and time consuming field trials.

The model also provides a tool with which to communicate with individuals in disciplines other than vector control. In the process of building the model, we created a multidisciplinary team including a naturalist from the park. As we developed the model it became quite apparent that each participant had gained an appreciation for the perspective of the other participants. A significant problem that we encounter in our preventive planning efforts is the inability of individuals in different disciplines to effectively communicate. Modelling may well be a means to bridge the problem if the process is accepted and used in land-use-planning.

The model is also useful in that it points out what we know, what we do not know, and the assumptions we have about the way the marsh operates. In the process of building the model, a great number of assumptions had to be made. For example, we had to make an assumption about the carrying capacity of the marsh for *Anopheles freeborni*, and the area searched in a given unit of time by mosquito fish, just to name two of the many assumptions. The process has provided a clear view of where we need additional information. Also, because the model is mathematical, it makes the kind of information required very specific. The model has therefore specifically defined the research that is required.

The model has also provided the benefit of showing clearly the logic involved in control decisions. The process of modelling has forced us to clearly document the logic which allows it to be evaluated and restructured as necessary.

Finally the modelling approach promises to dramatically change the role of the mosquito control technician. In the recent past in our district, the control technician was assigned a zone within which he or she had total responsibility for mosquito control, including making immediate treatment decisions involving extremely complex aquatic systems. The model will assist the technician in making these decisions and at the same time requiring more sophisticated and accurate sampling of the technician.

NEGATIVE IMPLICATIONS.–Although we foresee major benefits to be accrued from the model, there are also some problems that may arise and should be avoided where possible. Some of these problems are simply the other side of the coin of some of the benefits.

We are concerned that the model may actually block communication. Some people suffer from math and computer fear. For them the modelling process and the use of the model is not acceptable. If these individuals are in key positions the model has failed as a communication device.

Another problem is that the model tends to develop an aura of validity based upon the esoteric nature of diagrams, mathematics and computers rather than any actual reliability of the model in simulating the marsh. We must continually be testing the validity of the model and restructuring it as required. The logic and assumptions of the model should be available to all users for critical review.

The model also creates a problem in that its development requires the coordination and cooperation of a number of experts outside the district. Because we need help in the modelling process and because we are including more elements of the marsh in our considerations, we find ourselves dependent upon experts in modelling or in specific areas of aquatic systems.

Finally, the model requires that we upgrade our field sampling techniques. We must now find or develop effective methods to determine the number of mosquito fish that are present in the marsh at any given time. Standard dipping techniques will also probably have to be replaced by a more reliable system of sampling mosquito larvae.

CONCLUSIONS.–The use of the Coyote Hills Marsh Model promises to have a profound impact on the Alameda County Mosquito Abatement District. There is potential for many benefits that will be accrued during the development and use of the model. We are also concerned about possible pitfalls. We hope to enlist the help of others involved in similar endeavors to help us maximize the benefits and avoid the pitfalls.

ASSOCIATION OF *AEDES TRISERIATUS* AND *CULEX RESTUANS* IN WATER-FILLED TIRES

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ABSTRACT

Mosquito larvae were sampled in water-filled tires located within the New Jersey Pine Barrens in 1976 and 1977. Two types of tires were sampled–"AGED" tires (sampling period: 7/76-11/76, 5/77-9/77) which had been undisturbed for a number of years and "COLONIZING" tires (7/77-9/77) which were tires cleaned out and filled with litter and tap water.

The most common mosquitoes, *Aedes triseriatus* and *Culex restuans*, were found to be negatively associated (chi square = 36.5, $p < 0.001$, coefficient of association = -0.42) in the 1976 "AGED" tires. Commonly, a decline of *Ae. triseriatus* in a given tire was associated with the occurrence of *Cx. restuans* in the same tire. This species replacement in the "AGED tires was not observed in 1977; most of the "AGED" tires remained 100% or nearly 100% *Ae. triseriatus*. However, in the "COLONIZING" tires, there was a species succession of *Cx. restuans* to *Ae. triseriatus*.

Whether the negative association between these two species results from competitive interference or merely environmental heterogeneity is undetermined.

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SEASONAL AND SPATIAL DISTRIBUTION OF *TOXORHYNCHITES RUTILUS SEPTENTRIONALIS* RELATIVE TO ITS CULICID PREY IN A TIRE DUMP HABITAT

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ABSTRACT

The seasonal and spatial distribution of *Toxorhynchites rutilus septentrionalis*, a mosquito larvivore, was studied with respect to its culicid prey in a tire dump located within the New Jersey Pine Barrens during 1976 and 1977. This predator was not found until mid-July of the first year. Following an extremely harsh winter, it was not found during the second year until late August indicating that winter survival rate is low or zero in these tires. The seasonal range of the predator was shown to be very narrow as compared to its prey species, *Aedes triseriatus* and *Culex* species. This, along with asynchronous population peaks, caused seasonal overlap between the predator and prey species to be low.

The specific location of a tire within and around the tire dump proved influential in the spatial distribution of the predator. *Tx. r. septentrionalis* immatures were most abundant in tires leaning against trees. On the tire dump itself, they were most prevalent along the edge and decreased with increasing distance from the edge. The prey also demonstrated wider spatial distributions than the predator.

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MID-GUT AIR IN NEWLY EMERGED MOSQUITOES AND ITS ELIMINATION

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ABSTRACT

Air in the mid-gut of male mosquitoes is eliminated about one and a half hours earlier than that in females. Evidence is presented which suggests that the normal route is through the tissues.

INTRODUCTION.–That the mid-gut of newly emerged mosquitoes is filled with air is well known; Marshall and Staley (1932) described the phenomenon and showed how the oesophageal diverticula filled with bubbles of air whilst the air in the mid-gut was diminishing in volume. Indeed, they claimed that the mid-gut air passed forwards during the first few hours of adult life and was taken up by the diverticula, a view that has been widely accepted ever since. Brumpt (1941), however, regarded this to be unlikely on anatomical grounds, and Clements (1963), while noting the sequence of these events in the newly emerged adult, interpreted them with caution. His caution was well founded since Venard and Pensri Guptavanij (1966) clearly demonstrated that the diverticular air is not derived from the mid-gut air.

The mid-gut air,according to Venard and Pensri Guptavanij, normally passes posteriorly to pass out via the anus. It is, however, puzzling to see how they could maintain this view when they clearly demonstrated that the mid-gut air is also lost at about the same time even when the anus is sealed. To account for this difficulty they postulated that the disappearance of the mid-gut air when both mouth and anus are sealed must be by the uptake of oxygen by the insect while the nitrogen diffuses into the tracheae. The work described in the present paper indicates that this seems likely to be the normal route; it also shows that elimination is faster in males than in females.

MATERIALS AND METHODS.–The mosquitoes used were the LSHTM strain of *Aedes (Stegomyia) aegypti* (L.). Male and female pupae were washed and isolated "dry" in individual tubes, according to the method described by Gillett (1982), and stored at 30°C. The time of emergence was recorded to the nearest five minutes.

When newly emerged adult *Ae. aegypti* are exposed to carbon dioxide (CO_2) the gas rapidly enters the already partly inflated mid-gut and distends it still further. Indeed, CO_2 enters even when the alimentary canal is isolated in saline, provided there is air already present in the mid-gut. Isolated guts already distended with air have been observed to burst when CO_2 is passed over the saline medium (Gillett, 1982). This gas, therefore, was avoided as an anaesthetic, the mosquitoes simply being stunned by knocking the tube containing each insect against the soft cover of a book. During the anaesthesia that followed, each mosquito was placed in saline and dissected to expose the mid-gut and diverticula. The gut was examined for the state of inflation, after which CO_2 was passed over the saline to see if the gas entered the mid-gut or not. Ten preparations were made of each sex at 0-1, 1-2, 2-3,...,9-10 hours after emergence from the pupa.

RESULTS.–Before carrying out the main experiment a preliminary test was set up to establish the mode of entry of CO_2 in teneral adults. Forty newly emerged adults of each sex were divided into four groups of ten each; in group one the head was removed and the neck sealed with molten wax; in group two the anus was sealed with molten wax; in group three both neck and anus were sealed; in group four the insects remained intact as controls. When exposed to CO_2 the gas entered the mid-gut with equal rapidity in all three sealed groups. Nor could any difference by detected in the rate of entry between normal insects and any of the three experimental groups. On return to atmospheric air the excess gas in the mid-gut was lost almost as rapidly as it entered in all four groups.

In studying the normal duration of mid-gut air in the two sexes, no difference was found during the first three hours of adult life. Between three and four hours (at 30°C) the mid-guts of the males began to lose air, deflation being complete by five hours. In the females, on the other hand, deflation did not begin until the fifth hour and was not complete until eight hours (Table 1). The points at which one would expect to find half the males and half the females examined still holding some air in the mid-gut was approximately 4.5 and 6.5 hours respectively.

DISCUSSION.–Two points emerge from this work: the earlier elimination of the mid-gut air in males and the evidence for the route of elimination in both sexes. A hyperinflated mosquito loses the excess CO_2 when returned to air as rapidly as it gained it when exposed to CO_2. Since it makes no dif-

TABLE 1. Loss of mid-gut air in newly emerged adult *Aedes aegypti* at 30°C

Age (hours) post emergence	Males		Females	
	No. examined	No. with mid-gut air	No. examined	No. with mid-gut air
0 - 1	10	10	10	10
1 - 2	10	10	10	10
2 - 3	10	10	10	10
3 - 4	10	10	10	10
4 - 5	10	6	10	10
5 - 6	10	0	10	8
6 - 7	10	0	10	5
7 - 8			10	3
8 - 9			10	1
9 - 10			10	0

ference to the speed of entrance and exit of the gas whether the head or anus or both are sealed, it is evident that the gas passes directly through the tissues. Buck (1962) in discussing the rates of diffusion of oxygen (O_2) and CO_2 through insect tissue points out that the enormous difference owes its origin to differences in solubility of the two gases and not to differences in their diffusion coefficients; because of its high solubility CO_2 has 36x permeability constant of O_2 (Chapman 1969). I am suggesting, therefore, that the CO_2 passes through the tissues and via the tracheal system of the mosquito (into and out of the mid-gut) in the liquid phase. Since the CO_2 is lost through the tissues it seems reasonable to suggest that the mid-gut air leaves by the same route. That CO_2 is lost in less than a minute, whereas the mid-gut air takes 4.5 hours in males and 6.5 hours in females may be accounted for by the much greater solubility of CO_2 compared with that of O_2 and the even greater difference between the solubilities of CO_2 and nitrogen.

It is tempting to argue that the slower elimination of the mid-gut air in females is associated with their greater size and hence greater bulk of tissue through which the gases must pass. Preliminary trials, however, with undersized females (produced through deliberate over-crowding in the larval stage) showed them to be no different from their larger female companions, but this needs confirmation.

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A FIELD RELEASE OF RADIO-STERILIZED MALES TO SUPPRESS AN ISOLATED POPULATION OF *CULEX TARSALIS*[1]

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ABSTRACT

For the 1981 sterile-male release trial a new colony of *Cx. tarsalis* was established from the release site during the late autumn of 1980. In the spring of 1981 the colony was mass-reared, males were radio-sterilized and released into a native population at the onset of field emergence in Kern County. In total, 84,652 sterilized males were released.

The radio-sterilized males were uncompetitive for field females against field males. Radio-sterilization was not detrimental, since sterilized males derived from field-collected pupae were fully competitive under similar field conditions in 1979 and 1980. The loss of competitiveness could not be attributed to loss of fitness, since the survivorship estimates for the released males by mark-release-recapture methods were the same as that of males from the target population.

Loss of competitiveness was attributed to assortative mating which in turn was initiated with laboratory rearing and the process of colonization. The two types of females involved in the experiments, field and laboratory-reared, mated more frequently with males of their own genotype. The preferential mating began with the initial rearing in the insectary, since the mating behavior data of female offspring from field-collected females was intermediate between that of field and colonized females. Thus, females behaved differently from their native parents after only one generation of insectary rearing.

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STUDIES ON THE POTENTIAL ENVIRONMENTAL IMPACT OF THE HERBICIDE THIOBENCARB (BOLERO®)

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ABSTRACT

In laboratory tests the LC_{50}'s of thiobencarb to mosquitofish were ca. 3 ppm in static tests and 1.3 ppm under continuous flow-through exposures. In residue accumulation tests, bluegill sunfish concentrate thiobencarb into their tissues from water up to levels of 200X within 24 hours, but these residues decline with longer exposures; rinsing of treated fish with untreated water results in a rapid loss of the active ingredient. In static accumulation tests thiobencarb degraded in soil treated at the suggested use rate and there was no uptake by channel catfish. Field applications of 4 lb AI/acre of both EC and granular formulations resulted in maximum initial water residue levels of ca. 1 ppm; this caused some mortality (maximum 25%) of mosquitofish, but not enough to indicate a serious problem. Since such applications are only made during the early growing season the side effects were regarded as minor. It would be best for mosquito abatement districts to stock mosquitofish, on rice fields treated with thiobencarb, several days after the herbicide application, if possible.

INTRODUCTION.–In order to develop strategies for the implementation of Integrated Pest Management (IPM) of rice field mosquitoes it is essential to understand the relationships between all agents that will be applied to this habitat. The herbicide thiobencarb (S-(4- chlorphenyl) methyl diethylcarbamothioate) was sold experimentally in California during 1981 for early-season use (two leaf stage of rice plant) at a rate of four pound AI/acre. As if is anticipated that this herbicide will be used widely here in future years, studies were conducted on its potential environmental impact.

MATERIALS AND METHODS.–**Analytical Methods.** The analytical methods which were used were provided by Chevron Chemical Company (1979). In general, water samples were extracted with hexane, the combined hexane phases were concentrated and analyzed by gas-liquid chromatography (GLC) using an electron capture detector (ECD); soil and fish samples were blended with hexane and Na_2SO_4, filtered, "clean-up" on an alumina column and the aliquot containing the active ingredient was concentrated and analyzed by GLC-ECD.

Samples of water, soil and fish tissues were fortified at 1.0, 0.5 and 0.1 ppm, in tripicate, and the percent recoveries from each was determined.

Laboratory Toxicity to Mosquitofish. Toxicity tests were run against *Gambusia affinis* Baird and Girard under both static and flow-through conditions. The mosquitofish were reared in outdoor cement tanks. Acclimation to the test conditions was initiated at least two weeks prior to initiating a test. Mortality during acclimation period was less than 5%. Mixed sizes and sexes of mosquitofish were used in each test. In the first two static toxicity tests, technical thiobencarb (94.5% AI) was used; it was necessary to use acetone as a co-solvent (0.1 ml/liter) and stir for 16 hours in order to dissolve the technical herbicide. The same concentration of acetone was used in the controls. In the third static test, an emulsifiable concentrate (EC) formulation was used. In the fourth toxicity test, the EC formulation was also used but was applied as a continuous, flow-through exposure, using an all-glass proportional diluter. The concentrations of thiobencarb were monitored at the beginning and the end of each test using the analytical methods described above. Ten mosquitofish were placed in each test chamber in four liters of water and the loading rate never exceeded 0.8 grams fish/L. Mortality was determined by the lack of opercular movement and failure to respond to gentle probing with forceps. Fish were checked for treatment effects every two hours during the first eight hours of exposure and at 24 hours thereafter until termination of 96 hours. LC_{50} values were determined by probit analysis and the confidence limits were calculated using the Litchfield and Wilcoxon method (1949). Measurements of dissolved oxygen and pH were monitored continuously during the tests.

Fish Accumulation Studies. These studies were designed to comply with EPA requirements for registration of pesticides. Exposures were made under dynamic (continuous flow-through) conditions using bluegill sunfish, *Lepomis machro-*

chiris Rafinesque as well as under static conditions using channel catfish, *Ictalurus punctatus* Rafinesque.

For dynamic exposures bluegills were exposed continuously to five concentrations of thiobencarb, using an all-glass proportional diluter, for periods of 0, 24, 48, 72 and 96 hours. Fish were analyzed immediately after these exposures as well as after exposures for 96 hours followed by rinse periods with untreated water for 24, 48, 72 and 96 hours. At the end of each exposure or exposure-rinse period, the fish were sacrificed, frozen (-20°C or below) and later samples of whole bodies were analyzed for thiobencarb.

For static exposure, experimental tanks (32 x 25 x 12 in. each), containing 60 pounds Atwater sandy loam each, were treated with thiobencarb 8-EC to give a dose of four pounds Al/acre (based on surface area). Eight such tanks were allowed to age under aerobic conditions for 30 days and were then flooded and allowed to stand for an additional 30 days; fingerling channel catfish (5.0 to 12.0 cm S.L. and weighing 1.5 to 10 gm each) were then added (42/tank). Oxygen levels were maintained by gentle aeration. At intervals of 1, 3, 7, 10, 14, 22, and 31 days fish were collected, sacrificed, frozen (-20°C or below) and later samples of whole bodies, tissues and viscera were analyzed for thiobencarb.

Also, samples of the soil were taken on the day of treatment, after the 30 day aeration period, after the 30 day flooding period and at each interval when fish samples were taken; these were analyzed for the active ingredient of thiobencarb. In addition, water samples were taken on the first and the thirtieth day after flooding and at each interval when fish samples were taken; these also were analyzed for thiobencarb.

Field Studies. In May 1981, a field test (Reike rice field, Merced County) was conducted to measure the side-effects of thiobencarb when applied to a rice field at the recommended rate (4.0 pound AI/acre). The EC formulation was applied at 4.0 pound AI in 10 gallons water/ac and a granular formulation (10%) at 40 pounds/ac; all applications were by fixed-wing aircraft. The toxicity of these treatments to mosquitofish was monitored by placing ft^3 cages containing 20 fish each in the plots prior to the application. Also water and soil samples (hydrosoil) were collected before and at two hours and at 1, 2, 3, 5, 7, 13, and 21 days posttreatment. These were extracted and analyzed for thiobencarb in the laboratory. Other aquatic organisms were sampled pre and posttreatment by dipping, trapping (minnow traps) and by area sampling.

RESULTS AND DISCUSSION.—**Analytical Methods.** The recoveries of thiobencarb from samples of water, soil and fish tissues fortified with 1.0, 0.5 and 0.1 ppm are shown in Table 1. These were considered as adequated in order to carry out the toxicity and accumulation studies.

Laboratory Toxicity to Mosquitofish. The concentrations of thiobencarb sought and those found using various formulations and under both static and flow-through conditions are shown in Table 2.

In static toxicity tests, the 96 hour LC$_{50}$ of thiobencarb to *Gambusia affinis* was 3.1 ppm (Table 3) using technical material and 2.6 ppm using an EC formulation; however, when the fish were placed under continuous flow-through exposures, the LC$_{50}$ lowered to 1.3 ppm.

In toxicity tests, concentrations above 4 ppm were inacurate due to the limited solubility of the thiobencarb in

TABLE 1. Recovery of thiobencarb from water, soil and fish tissue samples fortified at 1.0, 0.5 and 0.1 ppm.

Sample	Concn. (ppm)	Average % Recovery[a]	Range	Lowest Detectible Limit (ppm)
tap water	1.0	98.7	98.1- 99.2	
	0.5	99.2	98.6-100.0	
	0.1	92.0	90.8- 96.0	0.005
soil[b]	1.0	96.4	95.8- 98.5	
	0.5	98.3	98.0- 98.7	
	0.1	97.0	96.2- 99.1	0.01
fish	1.0	99.1	98.8-100.0	
	0.5	99.6	99.0- 99.9	
	0.1	89.0	88.4- 92.2	0.01

[a] Analysis performed in triplicate

[b] Atwater sandy loam

water. Less than half of the initial concentration remained at 96 hour in the static tests. It is apparent that use of the EC formulation results in higher toxicity in static tests and that a continuous exposure (flow-through) produced the lowest LC_{50} value, as would be expected. In test chambers treated above 3.5 ppm, mortality was observed within 6 hours and by 48 hours was 80 - 90% of the final (96 hour) value.

In toxicity studies with carp, Ochiai and Kubota (1978) reported that all of the fish died within 14 days following a continuous exposure to 3.2 and 1.8 ppm of thiobencarb but 75% survived following an exposure of 1.0 ppm for 10 days.

Fish Accumulation Studies. The accumulation of thiobencarb from water into bluegill tissues following continuous flow-through exposure is shown in Table 4. It is apparent that bluegills can concentrate thiobencarb to levels of ca. 200X within 24 hours, but for continuous exposures longer than this there is a reduced residue level. When bluegills were exposed and then subjected to rinse periods with untreated water, there is a rapid loss of the active ingredient with time. The evidence shows that exposures to thiobencarb will not result in long-term residues in fish tissues.

In the static accumulation study, thiobencarb persisted in the soil without apparent loss during the 30-day aerobic aging period (Table 5); however, the active ingredient apparently, totally decayed during the first 30 days of flooding. It is not surprising, therefore, that no residues in fish were found. Thus under the required conditions of the EPA guidelines for pesticide registration, there is no indication of persistent residues in soil which could be accumulated by bottom-feeding fish.

Field Studies. The concentrations of thiobencarb in the field plots (Table 6) treated with the EC formulations averaged 0.9, 0.6 and 0.5 ppm at 24, 28, 48 and 72 hours, respectively; the same values for the granular treatments were 0.3, 0.4, and 0.5 ppm, at the same intervals (Table 7). The cumulative mortality of mosquitofish was 5% at 24, 25% at 48 and 25% at 72 hours for the EC formulation and 0% for 24, 2% at 48 and 10% at 72 hours for the granular application. One week after the treatment, additional cages of fish were placed in these plots and no mortality occurred. Thus, the EC resulted in higher initial water concentrations of the active ingredient and higher fish mortality. However, the mortality observed was not high enough to indicate a serious problem. It would be best for mosquito abatement districts not to stock rice fields with mosquitofish until several days following thiobencarb treatment; but if this is not practical, the mortality to be expected does not appear to be so great as to prevent the fish population from re-establishing itself.

The plankton (Table 8) was generally reduced (31-75%) during a two week period following the treatment but then populations began increasing. The benthos also showed effects: there was a 65% reduction of chironomid larvae but oligochaeta were not affected (Table 9). There was no visible reduction of nektonic organisms (Tables 9 and 10) however their numbers were too low to judge effects (presumably the low numbers were due to an earlier treatment of this field with parathion for control of tadpole shrimp).

In conclusion, there were some minor side effects following the thiobencarb treatment but such effects during the early part of the season are not of a serious nature.

ACKNOWLEDGMENT.–We thank the personnel of Wilbur-Ellis and Chevron Chemical Companies for allowing us to conduct the field studies on their experimental plots. This study was supported, in part, by EPA Cooperative Agreement No. CR-806771-01-1 and by a special California State appropriation for mosquito control research.

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TABLE 2. Concentrations of thiobencarb in chambers used for exposing mosquitofish under static and flow-through conditions.

Concn Sought (ppm)	Concn Found (ppm) 0 hrs	96 hrs	% of Initial Concn Remaining after 96 hrs
	Static Test No. 1-technical thiobencarb		
8.0	5.62	2.83	50.4
4.45	4.03	1.15	28.6
2.56	2.37	0.62	26.5
1.44	1.35	0.17	12.6
0.80	0.80	0.007	0.9
	Static Test No. 2-technical thiobencarb		
4.0	4.08	1.45	35.6
3.5	3.46	0.62	18.1
3.0	3.19	1.54	48.2
2.5	2.42	0.40	16.7
2.0	1.95	0.38	19.2
	Static Test No. 3-EC formulation		
4.0	4.90	1.98	40.4
3.5	3.95	1.50	40.0
3.0	3.00	1.15	38.3
2.5	2.50	0.78	31.2
2.0	2.25	0.18	8.0
	Flow-Through Test No. 1-EC formulation		
9.00	6.82	6.18	90.6
5.04	4.76	4.64	97.5
2.88	3.06	3.21	104.9
1.62	1.96	2.22	113.3
0.90	1.05	1.29	122.9

TABLE 3. Toxicities of thiobencarb to *Gambusia affinis* under static and flow-through exposures.

Test No.	Type Test and Formulation	Concn Used (ppm)	96 hrs LC_{50} (ppm)
1	Static-Technical	sought: 0.80 1.44 2.56 4.45 8.00 found: 0.81 1.35 2.37 4.03 5.62 LC_{50} confidence limits[a]: 3.71-2.59	3.10
2	Static-Technical	sought: 2.00 2.50 3.00 3.50 4.00 found: 1.95 2.42 3.19 3.46 4.08 LC_{50} confidence limits: 3.26-2.84	3.06
3	Static-EC	sought: 2.00 2.50 3.00 3.50 4.00 found: 2.25 2.50 3.00 3.95 4.90 LC_{50} confidence limits: 2.78-2.39	2.59
4	Flow-Through	sought: 0.90 1.62 2.88 5.04 9.00 found: 1.05 1.96 3.06 4.76 6.82 LC_{50} confidence limits: 1.55-1.17	1.34

[a] Determined by Litchfield-Wilcoxon method (1949).

TABLE 4. Residues of thiobencarb in bluegill tissues treated with continuous exposures and with continuous exposure-rinse periods (in ppm).

Exposure Period (hrs)	Rinse Period (hrs)	Treatment Concn[a] ppm 0.14	0.20	0.36	0.53	1.24
24	0	41.5	25.3	45.9	90.9	207.0
48	0	30.0	21.6	52.5	44.2	94.4
72	0	9.0	18.7	16.3	49.2	105.2
96	0	14.0	14.0	15.6	57.5	108.8
96	24	5.6	9.5	9.8	57.7	176.5
96	48	4.7	7.1	8.8	13.2	124.8
96	72	3.1	6.1	8.1	16.8	89.0
96	96	1.0	1.3	2.3	7.3	18.3

[a] Average daily concentration determined by chemical analysis.

TABLE 5. Thiobencarb static accumulation test-analysis of soil, water and channel catfish tissues.

	day	date	Soil (ppm)	%H_2O	H_2O (ppm)	Fish (ppm) whole body	edible tissue	viscera
soil treatment	0	7-13-81	0.3534		---	---	---	---
	30	8-12	0.4021		---	---	---	---
water added	31	8-13	---		0.031	---	---	---
fish added	60	9-11	ND[a]	20.8	ND[b]	---	---	---
	61	9-12	ND	21.2	ND	ND[c]	ND	ND
	63	9-14	ND	20.0	ND	ND	ND	ND
	67	9-18	ND	18.2	ND	ND	ND	ND
	70	9-21	ND	18.8	ND	ND	ND	ND
	74	9-25	ND	22.0	ND	ND	ND	ND
	82	10- 2	ND	21.2	ND	ND	ND	ND
	91	10-11	ND	18.0	ND	ND	ND	ND

[a] Less than 0.01 ppm - soil samples.

[b] Less than 0.005 ppm - water samples

[c] Less than 0.01 ppm - fish samples.

TABLE 6. Plot size and assignment of treatments of 4 lb AI thiobencarb to the Reike rice field.

Plot No.	Acre	Formulation
1	7.7	Control
2	7.0	EC
3	5.1	GR
4	7.5	EC
5	6.8	GR
6	10.3	Control
7	11.2	GR
8	11.8	EC
9	19.7	Control
10	19.7	EC
11	17.0	GR
12	20.6	Control

TABLE 7. Residues of thiobencarb in water and soil of a rice field treated with 4.0 lb AI/acre on 4/19/81 in Merced County, California.

Formulation:	EC				Granule			
Plot no:	2	4	8	10	3	5	7	11
Pre								
water	ND[a/]				ND			
soil	ND[b/]				ND			
2-hrs								
water	1.7	1.0	0.4	0.3	0.1	0.2	0.1	0.1
soil	0.3	0.1	0.2	0.2	0.1	0.01	0.4	0.3
1-day								
water	0.05	0.4	0.3	0.4	0.4	0.7	1.6	1.0
soil	0.04	0.02	0.06	0.08	0.03	0.2	0.2	0.05
2-days								
water	0.1	0.2	0.3	0.2	0.8	0.4	0.3	0.4
soil	0.2	0.2	0.2	0.1	0.2	0.2	0.1	0.1
3-days								
water	0.006	0.5	0.5	0.3	0.6	0.9	0.7	0.6
soil	0.7	0.9	0.6	0.6	0.8	0.8	0.6	0.5
7-days								
water	0.005	0.1	0.1	0.06	0.1	0.1	0.3	0.1
soil	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
13-days								
water	0.007	0.008	0.02	0.01	0.006	0.03	0.03	0.05
soil	0.2	0.2	0.4	0.6	0.2	0.06	0.2	0.8
21-days								
water	ND	ND	ND	ND	ND	ND	ND	ND
soil	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2

[a/] ND = not detected (<0.002 ppm).

[b/] ND = not detected (<0.01 ppm).

TABLE 8. Effect of thiobencarb on plantkon and benthos of the Reike rice field (in no. of organisms/20 dips).

	May						June	
Organism	18	20	21	22	26	27	1	9
plot no. 1			Control[a]					
Cladocera	18	11	9	17	10	13	6	30
Copepod	10	17	19	28	36	85	103	44
Ostracod	55	26	39	24	138	257	346	54
Mayfly	0	0	0	0	0	0	1	8
Corixid	0	0	0	0	2	1	2	3
Chironomid L	3	6	28	2	18	78	65	14
plot no. 2			EC					
Cladocera	69	3	0	3	1	6	14	13
Copepod	16	7	0	5	8	20	28	24
Ostracod	45	39	37	35	72	62	99	190
Mayfly	0	0	0	0	0	1	1	0
Corixid	0	0	0	0	0	1	0	0
Chironomid L	2	4	4	4	2	27	16	20
plot no. 3			Granules					
Cladocera	6	2	0	2	1	1	1	13
Copepod	10	8	13	25	4	14	3	82
Ostracod	24	28	30	133	19	128	240	254
Mayfly	0	0	0	6	1	1	1	3
Corixid	0	0	0	0	0	0	0	0
Chironomid L	2	15	13	19	4	0	3	5

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[a] Propanil was applied on June 4.

TABLE 9. Effect of thiobencarb on benthos and nekton of the Reike rice field (in no. organisms/3 area samples, 452 cm^2 each).

				May			June	
Organism	18	20	21	22	26	27	1	9
	plot no. 1			Control[a]				
Mayfly	2	2	1	3	0	0	0	9
Corixid	5	10	11	22	2	2	3	2
Hygrotus sp. L (A)	2	1	5(4)	4	4	4	4	3
Tropisternus sp. L	0	0	3	0	0	0	1	0
Chironomid L	279	305	294	392	264	584	945	75
Oligochaeta	4	1	3	9	15	18	12	26
	plot no. 2			EC				
Mayfly	0	0	0	0	0	0	0	0
Corixid	5	0	3	4	9	3	0	4
Hygrotus sp. L (A)	2	1	1	3	3	3	1	3
Tropisternus sp. L	0	0	0	0	2	4	1	1
Chironomid L	176	58	104	59	16	31	561	386
Oligochaeta	8	21	12	11	6	5	16	28
	plot no. 3			Granules				
Mayfly	0	0	0	0	0	0	0	0
Corixid	3	0	0	2	5	0	1	4
Hygrotus sp. L (A)	3	0	0	0	0	0	0	3
Tropisternus sp. L	0	0	0	0	0	1	0	0
Chironomid L	72	53	51	44	12	14	17	116
Oligochaeta	17	39	43	49	12	15	17	74

[a] Propanil was applied on June 4.
L = larvae, A = adults.

TABLE 10. Effect of thiobencarb on nekton associated with rice field (in no. organisms caught in 3 minnow traps in a 24 hr period).

	May			June
Organism	21	22	27	1[a]
	Control			
Mayfly	0	3	0	0
Corixid	15	40	11	6
Notonectid	2	2	0	0
Hygrotus sp. L (A)	1(1)	0	0(1)	0(4)
Laccophilus sp. L (A)	1(3)	0(5)	0(1)	0(7)
Tropisternus sp. O. (A)	0(9)	2(0)	0(5)	3(3)
Spider	0	0	5	14
Fish[b]	0	0	0	13
	EC			
Mayfly	1	0	0	1
Corixid	5	11	33	8
Notonectid	0	1	0	0
Hygrotus sp. L (A)	0	0	0(1)	0(17)
Laccophilus sp. L (A)	1(0)	0(1)	0(9)	0(8)
Tropisternus sp. L (A)	0(2)	0(2)	6(0)	4(9)
Spider	0	0	2	6
Fish[b]	1	3	0	3
	Granules			
Mayfly	0	0	0	0
Corixid	6	6	9	5
Notonectid	1	0	0	0
Hygrotus sp. L (A)	0(3)	0(1)	0(1)	0(9)
Laccophilus sp. L (A)	0(1)	2(0)	0(7)	0(6)
Tropisternus sp. L (A)	0(2)	0	1(3)	6(19)
Spider	0	0	2	2
Fish[b]	0	0	0	4

[a] Traps were left in the field for 5 days.

[b] Mostly carps and few mosquitofish.

L = larvae, A = adults.

FIELD TRIALS OF FICAM ULV® ON CAGED AND WILD MOSQUITOES IN CALIFORNIA[1]

R.M. Miller[2], P.A. Gillies[3], K.R. Townzen[3], R.K. Washino[4]

ABSTRACT

A 25.8% bendiocarb formulation was applied as an ultra low volume (ULV) aerosol to caged *Culex pipiens quinquefasciatus, Culex tarsalis, Anopheles freeborni* and wild populations of *Aedes nigromaculis* in Sutter and Butte Counties of California. Mortalities of 100% in caged mosquitoes were obtained at distances 3-5 times greater than the labeled swath width of 330 feet. Sustantial reductions of the wild mosquito population were also observed.

INTRODUCTION.–Ultra low volume (ULV) field trials performed during the past two years on caged mosquitoes have demonstrated bendiocarb to be an effective mosquito adulticide (Tapley et al. 1980, Atkins et al. 1981). Field trials were conducted in August 1981 on irrigated pastures in Sutter and Butte Counties of California to monitor the efficacy of this compound on caged mosquitoes and on the wild *Aedes nigromaculis* populations present at the test sites. These trials were designed to determine if previously published data on the mortality of caged mosquitoes are indicative of the effect of ULV-applied bendiocarb on wild mosquitoes.

MATERIALS AND METHODS.–Three separate field trials were run at different locations during two evenings to monitor the effects of Ficam ULV insecticide on caged and wild mosquitoes. Wild mosquito population densities were assessed by landing counts in all tests. The deZulueta trapping method was utilized in test three. Caged mosquitoes were placed in test lines running perpendicular and downwind from the path of the aerosol generator. Ficam ULV was supplied by the BFC Chemicals, Inc., 4311 Lancaster Pike, Wilmington, Delaware 19805. All tests utilized a truck mounted LECO HD aerosol generator equipped with a MICRO-GEN DFK-L digital flow control system. The aerosol nozzle was directed at a 45° angle away from the ground and the flow rate was determined to be accurate immediately prior to each evening trial. All tests were conducted using a vehicle speed of five miles per hour.

Meteorological data were compiled every fifteen minutes commencing one hour prior to the trials and terminating after release of the aerosol. Temperature was monitored at two meters and ten meters above the ground with an electronic thermistor thermometer. A hand held anemometer was used to measure wind speed and direction. A stability ratio was calculated according to a formula adapted from Haugen et al. (1961) as follows:

where

$$\frac{t_2 - t_1 \times 10^5}{(u)2}$$

t_2 = temperature °C at 10 meters
t_1 = temperature °C at 2 meters
u = average wind speed (cm/sec)

Field trials were initiated only when a temperature inversion was detected.

Three species of known susceptible adult mosquitoes were utilized for the cage tests. Species used included: *Culex pipiens quinquefasciatus* from the colony maintained by the California Department of Health Services, Vector Biology and Control Branch Laboratory at Sacramento; and *Culex tarsalis* and *Anopheles freeborni* from colonies maintained at the University of California, Davis, Department of Entomology. Approximately twenty unsexed adult mosquitoes of one species were placed in each disposable cage. Grade stakes were placed in a downwind line across the length of the test pastures. The caged mosquitoes were affixed to stakes three feet above the ground. Disposable cages were constructed of cardboard tubing and nylon netting as described by Townzen and Natvig (1973). The cages were left in the test fields for fifteen to thirty minutes following the ULV application to ensure that the aerosol had passed through the entire test line. Exposed cages were placed in individual plastic bags, moisture pads were provided for the mosquitoes and they were returned to the laboratory for processing.

Controls utilizing the species involved in the field test were placed in an adjacent pasture well up wind from the path of the ULV aerosol generator. They were retrieved after the collection of the exposed cages by uncontaminated personnel.

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[1] The opinions and assertions contained herein are the private ones of the writers and are not to be construed as official or reflecting the views of the Navy Department or the naval service at large.

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The controls were maintained in the same manner as the exposed mosquitoes during the posttreatment monitoring period.

Two or more investigators performed pre- and post- treatment landing rate counts of the wild field mosquitoes by walking across different areas of the test field. Counts began near the aerosol discharge line and extended downwind across the pasture. Each investigator stopped periodically as he traversed the test pasture and counted the number of mosquitoes on one pant leg. Pretreatment landing counts were performed twelve hours and immediately prior to each trial. Posttreatment landing counts were taken approximately twelve hours after each ULV application. Pre- and post-treatment landing counts separated by twenty-four hours were utilized to determine *Ae. nigromaculis* reduction because of the periodicity of the species (Miura 1971).

DeZulueta trapping, as described by Washino et al. (1977), was utilized in addition to landing counts to monitor wild mosquitoes in trial three. The trapped mosquitoes were collected and processed by personnel from the Entomology Department, University of California, Davis.

Test one was conducted on a 240 acre pasture in Sutter County. Caged *Cx. p. quinquefasciatus* were stationed at 0′, 150′, 300′, 450′, 600′, 800′, 1000′, 1200′, 1400′, 1800′, 2200′, 2600′, 3000′, 3400′, and 3675′ across the length of the pasture. A three ounce per minute insecticide flow rate was utilized. Landing counts were used to assess the wild mosquito population. Results of caged mosquito mortality and wild mosquito landing rates are given in the Tables 2 and 3 respectively.

Test two was performed on an 80 acre pasture in Sutter County approximately one and one half miles east and south of pasture one. Caged *Cx. p. quinquefasciatus* were stationed 0′, 150′, 300′, 450′, 600′, 800′, 1000′, 1200′, 1400′, 1600′, 1760′ from the path of the aerosol generator. A three ounce per minute insecticide flow rate again was utilized. Landing counts were used to assess the wild mosquito population. Results of caged mosquito mortality and wild mosquito landing rates are given in Tables 4 and 5 respectively.

Test three was run on an 80 acre pasture in Butte County. The southern side of this pasture was depressed and the road along the margin was elevated on an earthen bank three to four feet above the grass. Caged *Cx. p. quinquefascitus*, *Cx. tarsalis*, and *An. freeborni* was suspended in separate cages at 0′, 150′, 300′, 450′, 600′, 750′, 900′, 1050′, and 1125′ across the length of the pasture. The insecticide was applied at the rate of two ounces per minute. Landing counts and deZulueta trapping were used to monitor the wild mosquito population. Results of caged mosquito mortality, wild mosquito landing rates and deZulueta trapping are given in Tables 6, 7 and 8 repectively.

Pertinent data as to the date, time, duration, discharge rate, temperature, wind speed, wind direction and air stability ratio for each test are listed in Table 1.

RESULTS AND DISCUSSION.–This field work was performed under ideal climatic conditions. The wind was constant and averaged in speed from 1-2 miles per hour. A strong temperature inversion was present during each test. Field test one achieved 100% mortality of caged *Cx. p quinquefasciatus* between 150 feet and 1400 feet (Table 2), and a 75.8% reduction of wild *Ae. nigromaculis* (Table 3) as assessed by landing counts. Field test two achieved 100% mortality of caged *Cx. p. quinquefasciatus* through 1760 feet (Table 4) and a 98.4% reduction of wild mosquitoes (Table 5). Field test three achieved 100% mortality of caged *Cx. p. quinquefaciatus Cx. tarsalis* and *An. freeborni* between 150 feet and 1050 feet (Table 6). Landing counts indicated an 82.9% reduction of *Ae. nigromaculis* (Table 7) and the deZulueta trapping demonstrated a 96.5% reduction (Table 8).

Caged mosquito mortality in tests one and three were low at zero feet but increased to 100% at the second station and for a considerable distance down the test line in each test. This information indicates the aerosol passed over the mosquitoes in the cages at zero feet while further downwind the insecticide settled due to the strong temperature inversions present. Field test three supports this premise with the deZulueta trap data indicating a high reduction of *Ae. nigromaculis*. The deZulueta trapping was performed at random approximately 500 feet from the path of the ULV generator where the insecticide would be expected to settle. In situations such as these, a double nozzle aerosol generator with nozzles directed at different angles would be expected to treat the area adjacent to the insecticide discharge path more thoroughly than a single nozzle machine.

In all trials mortality patterns in caged mosquitoes indicate an effective swath width three to five times greater than would be expected from label information. Wild mosquito mortalities in tests one and three seem low but it must be assumed posttreatment counts were influenced by an influx of mosquitoes from adjacent fields. The results of these trials support data of Tapley et al. (1980) and Atkins et al. (1981), and indicate the bendiocrab ULV formulation utilized shows considerable promise as an effective mosquito adulticide.

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TABLE 1.

Resume of technical data for FICAM ULV® tests Sutter & Butte Counties

California, 1981.

PARAMETERS	Test 1	Test 2	Test 3
Date	8/24/81	8/24/81	8/25/81
Time	1920	2010	1920
Duration (min)	15	9	8.2
Discharge (fl oz/min)	3	3	2
Temp at 10m (°C)	24	22	26.5
Temp at 2m (°C)	21.2	18.5	21.8
Wind speed (mph)	1.5	1.0	2.0
Wind direction	SE	SE	SE
Air stability ratio	ca 40	100+	30+

TABLE 2.

Test 1: Percent mortality of caged *Culex pipiens quinquefasciatus* following aerosol application of Ficam ULV®, August 24, 1981.

Distance (ft)	Morality at intervals after exposure 12 HR	 24 HR
0	0	0
150	100	--
300	70	100
450	100	--
600	36	100
800	0	100
1000	21	100
1200	11	100
1400	17	100
1800	4	92
2200	6	94
2600	0	27
3000	0	70
3400	4	19
3675	0	53
Control		
1	0	0
2	0	0

TABLE 3.

Test 1: Percent reduction of *Aedes nigromaculis* twelve hours following aerosol application of FICAM ULV® , August 24, 1981.

Stop	$\bar{x}$ of *Ae. nigromaculis* per pant leg Pretreatment	 Posttreatment	 % Reduction
1	7.5	1	86.7
2	7.5	2	73.3
3	8	1	87.5
4	11	3	72.7
5	14.5	2.5	82.8
6	6.5	2	69.2
7	9	2	77.8
8	11.5	3	73.9
9	9.5	1.5	84.2
10	5.5	3.5	36.4
Mean All Stops	9.1	2.2	75.8

TABLE 4.

Test 2. Percent mortality of caged **Culex pipiens quinquefasciatus** following aerosol application of FICAM ULV®, August 24, 1981

Distance (ft)	Mortality at intervals after exposure		
	2 HR	12 HR	24 HR
0	100		
150	100		
300	85	100	
450	53	100	
600	5	100	
800	0	100	
1000	30	100	
1200	0	100	
1400	38	100	
1600	38	88	100
1760	21	100	
Control			
1	0	0	54
2	0	0	0

TABLE 5.

Test 2. Percent reduction of **Ae. nigromaculis** twelve hours following aerosol application of FICAM ULV®, August 24, 1981.

Stop	$\bar{x}$ of **Ae. nigromaculis** per pant leg		
	Pretreatment	Posttreatment	% Reduction
1	54	2.5	95.4
2	156	2	98.7
3	160	2.5	98.4
4	115	0.5	99.6
5	170	2	98.9
6	235.5	2.5	98.9
7	245	1.5	99.4
8	232.5	5	97.8
9	215	3.5	98.4
10	218.5	4	98.2
11	102	3.2	96.9
Mean All Stops	173.9	2.7	98.4

TABLE 6.

Test 3: Percent mortality of caged mosquitoes by species at intervals after exposure following aerosol application of FICAM ULV®, August 25,1981.

Distance (ft)	Percent Mortality					
	C. p. quinquefasciatus		**C. tarsalis**		**An. freeborni**	
	2 HR	12 HR	2 HR	12 HR	2 HR	12 HR
0	7	7	64		18	55
150	100		100		100	
300	100		100		100	
450	100		100		100	
600	INVALIDATED..					
750	INVALIDATED..					
900	100		100		100	
1050	100		100		100	
1125	INVALIDATED..					
Control						
1	0	0	20	10	0	0
2	0	0	18	18	0	18
3	0	0	0	0	13	0

TABLE 7.

Test 3: Percent reduction of **Aedes nigromaculis** twelve hours following aerosol application of FICAM ULV® , August 25,1981.

Stop	$\bar{X}$ of **Ae. nigromaculis** per pant leg		
	Pretreatment	Posttreatment	% Reduction
1	110	20	81.8
2	125	40	68
3	175	75	57.1
4	250	60	76
5	350	47.5	86.4
6	250	30	88
7	175	25	85.7
8	107.5	12.5	88.4
9	200	7.5	96.3
10	175	10	94.3
Mean All Stops	191.8	32.8	82.9

TABLE 8.

Test 3: Percent reduction of Aedes nigromaculis per deZulueta trap (24 ft^2) collection twenty-four hours following aerosol application of FICAM ULV®, August 25-26, 1981.

	Number of Ae. nigromaculis per 24 ft^2		
Collection	Pretreatment	Posttreatment	% Reduction
1	993	29	97.1
2	394	9	97.7
3	302	12	96
4	166	8	95.2
5	204	1	99.5
6	358	12	96.6
7	472	39	91.7
8	460	18	96.1
9	432	15	96.5
10	342	3	99.1
Mean All Stops	412.3	14.6	96.5

CONTROL OF IMMATURE MOSQUITOES WITH LIQUID AND SOLID FORMULATIONS OF A MONOMOLECULAR ORGANIC SURFACE FILM[1,2]

R. Levy[3], C.M. Powell[3], W.D. Garrett[4] and T.W. Miller Jr.[3]

ABSTRACT

Research and development of liquid and solid formulations of the monomolecular organic surface film ®Arosurf 66-E2 for mosquito control are discussed, with emphasis on their potential implementation in operational mosquito control programs.

In 1977 the Lee County Mosquito Abatement District initiated an applied research and development program aimed at evaluating alternative techniques for currently used conventional practices. The main objective was to determine if certain biological control agents and non-toxic chemicals could be integrated into our operational program and thereby reduce the use of conventional toxicants as well as solve specific operational problems that were characteristic to our area.

Since the onset of our program we have conducted research in areas of parasite, predator, and pathogen field efficacy and application technology as well as in mass production and storage. Several of these biological control agents are mass-reared at our district and are expected to become an integral part of our operational program.

One on-going reasearch project has involved the extensive evaluation of non-petroleum monomolecular organic surface films as potential larvicides and pupicides. The dramatic escalation in the cost of petroleum oils used for larviciding and pupiciding as well as the reported phytotoxicity and adverse effects on selected non-target organisms by diesel-based larviciding oils were the initiating factors for this research. Since the onset of the project we have evaluated over 50 types of organic surface films in the laboratory and field and have determined that one surface active chemical, i.e., isostearyl alcohol containing two oxyethylene groups (designated ISA-20E) is cost/effective and can be an acceptable alternative to the use of petroleum based oils as well as other larvicides used in mosquito control (Levy et al. 1980a, b; 1981; 1982a, b).

ISA-20E LIQUID FORMULATIONS.–ISA-20E is manufactured as a cosmetic ingredient by Sherex Chemical Company under the trade name of ®Arosurf 66-E2[5]. The efficacy of this product in mosquito control as well as the effectiveness of the oleyl alcohol indicator oil[6] to monitor field persistence of ISA-20E have been demonstrated at several mosquito control districts in Florida as well as by mosquito control agencies in other areas of the United States and Overseas. ISA-20E is currently being evaluated by ground and helicopter application on an operational basis at certain mosquito control districts in Florida under a permit granted to the Office of Entomology, State of Florida, by the Department of Environmental Regulation for use of ISA-20E as a larvicide and pupicide. Material cost per acre for ISA-20E at typical application dosages of 0.2-0.4 gal/acre is estimated at $2.50-5.00. Control of pupae can be accomplished at a rate of 0.1-0.2 gal ISA-20E/acre at an estimated per acre cost of $1.25-2.50. These costs are presently cheaper than typical mosquito control application rates of the Florida diesel-based larviciding formulation and FLIT MLO®, and are competitive with several other chemical and biological control agents used to control immature mosquitoes. It should be noted that higher dosages of ISA-20E, i.e., 0.75-1.0 gal/acre +, may also be cost/effective when used in dynamic permanent water *Culex* and *Anopheles* spp. habitats where highly prolonged film persistence (hence mosquito control) would be desirable. In addition, overdosing can be an acceptable procedure in certain situations where lateral expansion of semi-permanent mosquito habitats (eg. *Aedes* and *Psorophora* spp.) due to persistent rainfall is expected to occur within a few days after initial surface application. In this case, high mosquito-controlling film pressure can be maintained in the newly flooded breeding areas without reapplication of the material.

The larvicidal and pupicidal action of this surface film have been shown to be physical and not toxic in nature, being

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[1] Presented at the Joint Meeting of the American Mosquito Control Association and California Mosquito and Vector Control Association, April 18-22, 1982, Sacramento, California.

[2] Mention of a brand name or proprietary product does not constitute a guarantee or warranty by Lee County Mosquito Control District, and does not imply its approval to the exclusion of other products that may also be suitable.

[3] Lee County Mosquito Control District, P.O. Box 06005, Fort Myers, Florida 33906.

[4] Environmental Sciences Division, U.S. Naval Research Laboratory, Washington, D.C. 20375.

[5] Sherex Chemical Company, Inc., P.O. Box 646, Dublin, Ohio 43017.

[6] Manufactured as a cosmetic and pharmaceutical ingredient under the trade name of ®Adol 85 N.F. by Sherex Chemical Company, Inc., P.O. Box 646, Dublin, Ohio 43017.

related to habitat surface tension reduction and subsequent wetting of tracheal structures leading to suffocation. Therefore, resistance of mosquitoes to ISA-20E is not expected to develop.

Field trials at the Lee County Mosquito Control District have indicated that ISA-20E can be applied by conventional ground and aerial spray equipment as technical chemical at dosages ranging from ca. 0.20-0.50 gal/acre or at these recommended dosages as an agitated ISA-20E water-based suspension at final application rates of 5-7 gal/acre to effectively control larvae, pupae, and emerging adults of *Aedes*, *Anopheles*, *Culex*, *Psorophora*, and *Uranotaenia* spp. with little or no adverse environmental effects.

It should be noted that when ISA-20E is suspended in water with agitation, it is recommended that the system be thoroughly cleaned of petroleum oils (eg. diesel) and conventional pesticide residues prior to introduction of the ISA-20E. Diesel plus ISA-20E plus water can produce a milky gelatinous unsprayable material. ISA-20E can become contaminated with residues of pesticides left in the spray tank from prior use which may result in undesirable environmental and non-target stress. Spray systems can be cleaned with certain solvents (eg. 2-propanol) or with multiple washings of ISA-20E-water suspension. Detergents (soaps) are excellent for cleaning ISA-20E from hands, clothing, etc., but are not recommended for cleaning ISA-20E from spray tanks. Detergent residues remaining in the tank can act to destroy the film-forming-properties of ISA-20E and will therefore affect normal field persistence and spreading/respreading characteristics of ISA-20E.

Vigorous agitation of ISA-20E in the water will assure a homogeneous suspension and accurate application rates. Conventional paddle agitation and agitation by pump recirculation have been shown to produce effective results in truck and helicopter spray systems, respectively. It should be noted that the addition of a 30 gal/min submersible pump in a Bell 206 helicopter spray tank was shown to significantly improve agitation over the use of the normal 10 gal/min recirculation pump by-pass rate.

Additional research by Hertlein et al. (unpublished) has also shown that recommended dosages of ISA-20E can be injected into a stream of water for application at high spray pressures and volumes with the use of commercially available injection valves. It is expected that these new application procedures will greatly increase the range of effectiveness of ®Arosurf 66-E2 in the mosquito control programs.

ISA-20E entrapment of ovipositing females and resting males has been demonstrated in the laboratory and observed under field conditions. The sinking and inhibition of eclosion of egg rafts of *Culex* spp. have also been demonstrated in bioassays as well as under field conditions with the use of high pressure spray equipment. Therefore, under certain situations monomolecular films of ISA-20E can exert an ovicidal, larvicidal, pupicidal, and/or adulticidal effect on natural mosquito populations.

The larvicidal action induced by this surface film of 90% or greater was observed to occur within 24 hr post-treatment (i.e., acute kill), however, under most field conditions this level of mortality was usually obtained within 48-72 hr post-treatment (i.e., delayed kill). For the most part, the rate of mortality was attributed to species, instar, stage of development, habitat oxygen levels, habitat surface characteristics, and climatological factors. High pupal mortality usually occurred 2-4 hr after treatment. Prolonged persistence of ISA-20E at high film pressure under most environmental conditions was shown to produce the sustained physical impact on immatures when an acute larvicidal effect was not achieved. ISA-20E has been reported to persist in a variety of natural habitats from 2-10 days at recommended dosages. However, at any dosage, sustained wind fetch was observed to adversely affect application and uniform water surface coverage by translocating and compacting the ISA-20E in downwind areas. This was compounded when high concentrations of floating organic and inorganic debris and vegetation was present in the habitat. When wind velocity was high and directional for extended periods, respreading of ISA-20E to displaced areas could not occur and subsequent control of immatures in locations of little or no film pressure was poor. For the most part, this was not a significant problem under conditions of fluctuating wind speed and direction; however, a delayed larvicidal response was usually observed. Conditions such as drainage, overflow and runoff have also been shown to adversely affect film performance in certain habitats.

ISA-20E SOLID FORMULATIONS.–Since mortality of natural populations of larvae exposed to ISA-20E could be delayed and was therefore a function of film persistence, research was conducted to determine if a solid biodegradable matrix could be developed for the release of monomolecular films of ISA-20E for the maintenance of high film pressure on immature mosquitoes for prolonged periods and thereby compensate for the limiting effects of persistent wind, runoff, and overflow. To effect the desired dispersal mechanism, ISA-20E was formulated with several dispersants, binders, and stabilizing components. The resultant solid/semi-solid matrices were evaluated under laboratory and field conditions against larvae and pupae of *Anopheles*, *Culex*, *Aedes*, and *Psorophora* spp. to determine if a stable mosquito-controlling monomolecular film could be consistently released for extended periods. The following set of criteria were the basis of evaluating the performance of over 25 solid formulations. An effective solid formulation of ISA-20E should: (1) Be composed of non-ionic and non-reactive components for use in a variety of water quality situations; (2) release surface films for prolonged periods under a wide range of temperatures; (3) release surface films for prolonged periods after exposure to drying and reflooding conditions; (4) maintain a solid consistency and release minimal dosages after long-term field exposure; (5) release monomolecular films upon contact with water with little or no delay; (6) have a persistent larvicidal and pupicidal effect against a wide variety of mosquito species; (7) be non-toxic, biodegradable, and produce little or no adverse environmental impact after prolonged field use; (8) be capable of being fabricated into beads, pellets, blocks and cylinders for a variety of application techniques; (9) have a stable shelf-life after prolonged storage at ambient temperatures; (10) and be composed of materials that are commercially available at a reasonable cost.

Initial field trials were conducted with three floating solid formulations containing 30-50% ISA-20E that were anchored in an upwind location in 70 m^2 standing paludal ponds containing various amounts of floating and emergent vegetation. Results of these tests against mixed natural populations of *An. quadrimaculatus*, *An. crucians*, and *Cx. erraticus* indicated that ISA-20E could be released from a 280-290g biodegradable matrix for over five months under field conditions and control 90-100% of the immatures. It should be noted that weekly spray treatment of ISA-20E at a dosage of 0.4 gal/acre was required to achieve this level of control. Although prolonged film release with the solids were demonstrated, the matrices softened too rapidly and released an excess amount of surface film. Therefore, these formulations were not suitable for operational mosquito control. In cooperation with Sherex Chemical Company, we have greatly improved the consistency of the matrix and the rate of film release. To date, over 25 solids have been evaluated in the laboratory against *Culex* and *Aedes* spp. to determine which formulations adequately meet the criteria we have established for a effective solid.

Laboratory tests against larvae and pupae of *Ae. aegypti* with several 0.5-7 g solid formulations containing 40-50% ISA-20E have indicated that a stable and effective mosquito-controlling surface film can be maintained in containers for 2-5 months and produce cumulative mortality of larvae, pupae, and emerging adults of 90-100%. Similar results were obtained in tests against *Ae. taeniorhynchus* and *Cx. quinquefasciatus*. The entrapment of ovipositing females of *Cx. quinquefasciatus* on surface films of ISA-20E released by the solids has also been demostrated.

Preliminary field tests in salt-marshes against *Ae. taeniorhychus* with 5-15 g cubes and 280-290g cylinders of these formulations indicated that monomolecular films of ISA-20E can be released for extended periods to control larvae and pupae in semi-permanent habitats. Results indicated that certain formulations are stable enough to release surface films after being subjected to periods of drying and reflooding. Similar results were obtained in tests against *Ps. columbiae* and *Ps. ciliata*. It should be noted that the effective use of the indicator oil in monitoring the release of ISA-20E from solid formulations and its persistence in a mosquito habitat have been demonstrated. Although the results of tests with these formulations are extremely promising and indicate their potential use in certain situations for season-long mosquito control from one application, problems concerning matrix dissociation after prolonged exposure to water have to be solved before reliable formulations can be developed for use in operational programs. Field trials with new solids are scheduled for this summer. It should be noted that ISA-20E intermittent drip-dispensing systems can be used to maintain persistent film pressure for several months in habitats such as paludal ponds and irrigation and sewage treatment systems.

In summary, research at the Lee County Mosquito Control District has shown that ®Arosurf 66-E2 is a safe and effective product for use in mosquito control programs. Solid formulations of this monomolecular surface film are expected to greatly increase the range of its effectiveness as well as reduce application costs.

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COMMITTEE ACTIVITIES OF THE CALIFORNIA MOSQUITO
April, 1981-April, 1982
AND VECTOR CONTROL ASSOCIATION, INC.

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This past year was very active for many of the Committees of the California Mosquito and Vector Control Association, Inc. The success of any Association is normally equal to the amount of effort put forth by its membership and the CMVCA is a prime example of a successful association due to its firm committee foundation.

The following is a review of the major accomplishments and activities of the various committees for 1981-1982 year.

BUDGET AND EXECUTIVE.–The Budget and Executive Committee is charged with preparing the Agenda for the Board of Directors' Meetings, preparation and analysis of the budget, and the coordination and stimulation of CMVCA activities.

Although the Committee accomplished many tasks during the year, the single most important accomplishment was the establishment of a salary and benefit package for the Association Office Secretary.

COMPUTER COMMITTEE.–This Committee's charge is to maintain communication and coordinate the program activities of mosquito abatement districts which may acquire a computer; develop a style manual for programming; develop a communications program; update the CMVCA program disc; and provide training by personal contact and/or seminars or workshops.

During the past year, the Committee set up a computer program for registration at the CMVCA-AMCA Annual Conference. They plan to update the CMVCA computer disk and have renamed it "CMVCA Computer Committee Resource Disk". A style manual is currently under development and review by the Committee.

CONTROL COMMITTEES.–**Biological Control.** The Committee is currently working on a continuation of Bio Briefs with Gary Reynolds of OCVCD as Editor and the preparation of a series of one-page notes on biological control agents other than fish. The Committee held a one-half day workshop in conjunction with the S.O.V.E. in January, 1982. The workshop emphasized new findings in mosquito control using fish and operational implementation of these finding.

Chemical Control. Kept abreast of chemicals currently being used for mosquito control.

Physical Control. The Committee discussed the prevention and control of mosquitoes in dairy waste ponds, specifically manure separation systems; weed control and maintenance and floating matter; agricultural land subdivision; and underground utility vaults. Recommendations on these matters will be forthcoming.

COOPERATIVE PLANNING COMMITTEE.–The Cooperative Planning Committee working with the Environment Committee has created the beginning of a data base of rare and endangered species that could be affected by vector control activities. A computer program has been created by the Committees to allow vector control personnel access to the data base in order to detect any potential impact on rare or endangered species by any of the vector control methodologies. The Committee met with wildlife representatives, and they all concluded that a computerized approach to detecting potential environmental problems resulting from vector control is appropriate; the system should be expanded where possible to include local information about distribution of rare and endangered species and that the system cannot replace the important personal contact that should be established between vector control agencies and wildlife personnel.

ENTOMOLOGY COMMITTEE.–The Committee co-sponsored along with the Biological Control Committee and S.O.V.E. our Annual Conference held in January of 1982.

ENVIRONMENT COMMITTEE.–This Committee has been working closely with the Cooperative Planning Committee this year on development of a computer program for evaluation of the impact of vector control activities on rare and endangered species.

EQUIPMENT COMMITTEE.–The charge of this Committee is to study various types of control equipment and report to the membership on important innovations or developments. However, this year they were primarily involved with equipment used by the central office, principally printing equipment and accessories. They have done an outstanding job of evaluating the prospective purchases of the Association in regards to printing equipment.

LEGISLATIVE COMMITTEE.–This Committee has performed an outstanding service to the Association by keeping the members abreast of legislative developments and working closely with the Association's legislative advocate. Many legislative bills were reviewed with some of special interest to the CMVCA. Senate Bill 618, Hazard Waste Control: we are attempting to amend the bill to exempt vector control

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[1] Vice President, California Mosquito and Vector Control Association, Inc.

agencies that are signatory to the cooperative agreement from the fees and monthly reporting requirements. A vector control funding bill AB 1662 did not make it out of Committee and was filed in the inactive two-year file. SB 65, the Emergency Invasion bill involving catalitic converters on vector control agencies vehicles, passed the legislature exempting vector control agencies from the requirement of having catalitic converters on spray vehicles.

LOCAL ARRANGEMENTS AND PROGRAM COMMITTEES.–Both Committees worked in concert to locate the site and develop the program for the 50th Annual Conference of the CMVCA in a joint meeting with the AMCA at the Capitol Plaza Holiday Inn, Sacramento, California, April 18-22, 1982.

PUBLICATIONS COMMITTEE.–This committee spent much time and effort in getting the printing operation going again by conferring with the central office personnel and the Secretary, lending their expertise and valuable advice. The Committee edited the papers for inclusion in the 49th Proceedings and recommended that the 45 days given authors to submit their papers for publication be ridgedly enforced. The Committee also worked on the preparation of the 1981-82 Yearbook, the publication of which was imminent.

RESEARCH COMMITTEE.–The Committee is charged with the determination of the research and technical developmental needs of California mosquito and related vector control. The Committee reviewed in depth the research proposals submitted by research scientists for funding via mosquito research funds provided by the State of California. Following their review of proposals, the Committee rates the proposals and makes recommendations to the various University Committees that review and make the final recommendations for moquito research fund allocations.

WAYS AND MEANS COMMITTEE.–This Committee worked most of the year on the new central office. The Committee recommended that Kathy Morrison be retained to train the new Office Secretary. After much discussion, recommended the purchase of a collator and installed same in the central office. The Committee was charged with studying the corporate member dues structure and making appropriate recommendations to the Board of Directors.

FISCAL INFORMATION COMMITTEE.–This Committee has been compiling fiscal data from quarterly reports submitted by each vector control agency. This information is to be used in dealing with the legislature in any new funding bills.

All of the Committees are to be commended for the outstanding accomplishments achieved this year.

PUBLICATION POLICIES AND INFORMATION FOR CONTRIBUTORS

"THE PROCEEDINGS" is the Proceedings and Papers of the California Mosquito and Vector Control Association, Inc. One volume is published each year. Intended coverage by content includes papers and presentations of the Association's Annual Conference, contributions and meritorious reports submitted for the conference year, and a synopsis of actions and achievements by the Association at large during the preceding year.

CONTRIBUTIONS: Articles are original contributions in the field of mosquito and related vector control providing information and benefit to the diverse interests in technical development, operations and programs, and management documentation. Papers on controversial points of view are accepted only as constructive expositions and are otherwise generally dissuaded, as is the case with an excessive number of papers on one subject or by one author where imbalance might ensue. Although preference is given to papers of the conference program, acceptability for publication rests on merit determined on review by the editors and the Publications Committee.

MANUSCRIPTS: The diversity of interests and fields of endeavor represented by contributors and readership of the Proceedings precludes strict conformance as to style. Authors should refer to recent issues of Mosquito News for general guidance. Authors of technical papers should follow the basic recommendations as presented in the Council of Biology Editors Style Manual. Authors should submit an original on white bond paper, with one additional copy. All parts of manuscripts (text, tables, references and legends) must be typed, double-spaced with ample margins. Avoid footnotes in text. Author should indicate with pencil, in the margins, the approximate positions desired for illustrations and tables.

The complete scientific name of an organism must be given the first time it is used. Terms commonly abbreviated in specific fields should be given in full the first time used, followed with the abbreviation. The abbreviation alone is acceptable in further usage in the paper. Common latin abbreviations (et al., e.g., i.e., etc.) are not italicized. Use of the metric system is encouraged. The bi-letter system of generic abbreviations is used for Culicidae.

All manuscripts will be edited to improve communications, if needed. Editors are biased against verbosity or needless complexity or jargon. Grammar will be corrected if necessary. Articles needing extensive editing or not conforming to style and instructions will be returned to the author for correction.

Manuscripts should be submitted to the editor within 45 days after the Annual Conference to ensure publication. Mail all material to the CMVCA PRESS, 197 Otto Circle, Sacramento, California 95822.

ABSTRACTS: Only technical papers need be accompanied by an abstract, which should not exceed 3% of the length of the article. When an abstract is submitted for publication in lieu of a paper, the abstract length may be extended. If possible, the journal where the paper is to be published in full should be stated.

TABLES: Tables are typed on separate sheets placed in correct sequence in the text and should be limited to those strictly necessary. Tables made up by the author in the form of line drawings for photocopy are acceptable. Graphs and line drawings should be prepared with regard to the ultimate printed size of one column (3¼") or page width of 7 inches, as applies to columns of table data.

Submitted figures as maps and charts should not exceed 8½ X 11" (22 X 28 cm), with labels and line weight adapted to the published size. Total page space for tables and figures must necessarily be limited by the editors.

ILLUSTRATIONS: Illustrative material must be mailed flat. A copy for use of reviewers is desirable. Figures should be numbered consecutively. Illustrations prepared for printing as line drawings are preferred but those requiring half tones are acceptable. Titles, legends or other headings should be grouped according to the arrangement of the figures and are typed double-spaced, on a separate sheet at the end of the paper. As with tables, the illustrations should be planned to fit reasonably the width of one column (3¼") or one page (7"). Figure numbers, as well as author's name and paper title should be written in blue pencil on the back of each illustration.

PROOF AND REPRINTS: Authors will receive page proof, as well as an order blank for reprints with a schedule of charges. Authors should not make major revisions of their work at this stage. Proofs with corrections, if any, should be returned within 10 days to the printer (CMVCA PRESS, 197 Otto Circle, Sacramento, California 95822.