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January 28-29-30-31, 1973

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SANTA BARBARA, CALIFORNIA

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## TABLE OF CONTENTS

The Legislative Outlook Relating to Mosquito Control . . . . .	Robert J. Beckus	1
EPA – Its Impact upon California Mosquito Control . . . . .	George W. Milius	2
The Impending Threat of VEE to California Based on the Mexican Experience of 1972 . . . . .	Telford H. Work	3
Emerging Concepts of Factors that Limit the Competence of <i>Culex tarsalis</i> to Vector Encephalitis Virus . . . . .	James L. Hardy and William C. Reeves	7
The Veterinary Outlook Concerning Encephalitis Prevention . . . . .	Charles H. Burger	11
Surveillance for Arthropod-Borne Viruses and Disease by the California State Department of Public Health . . . . .	Richard W. Emmons, Gail Grodhaus and Dario Cappucci	13
Legal Aspects of Mosquito Control . . . . .	Robert H. Peters	27
What's New in Source Reduction . . . . .	George R. Whitten	28
Obstacles Presented by the State and Regional Water Resources Control Boards . . . . .	Dwight C. Baier	30
How Compatible are Mosquito Control and Agriculture . . . . .	C. Brunel Christensen	31
District Viewpoints on the use of the Legal Approach . . . . .	J. Warren Cook	32
Viewpoint on Use of the Legal Approach to Mosquito Control . . . . .	W. Donald Murray	33
Results of Abatement Hearings . . . . .	Theodore G. Raley	34
Experiences in the Persuasion of Property Owners to Alter their Land or Water-Use Practices . . . . .	Eugene E. Kauffman	35
Legal Actions for Abatement of Mosquito Breeding in the Southeast Mosquito Abatement District . . . . .	Gardner C. McFarland	36
A Planned Public Information Program . . . . .	Stephen M. Silveira	37
Student Concern with the Environment . . . . .	William H. Steinmetz	38
Trustees' Concern over Limitations of Pesticide Use . . . . .	William S. Brown	39
Pesticides Can Be Compatible with the Environment . . . . .	John E. Swift	41
Future Pesticide Methods When Chemical Uses Are Curtailed . . . . .	E. J. Dietrick	42
Environmental Approaches to Vector Control . . . . .	Richard F. Peters	43
Responsibilities and Services to Mosquito Abatement Districts . . . . .	Boysie E. Day	45
State Agencies Concerned with Mosquito Control . . . . .	Frank M. Kozlik	46
Fish and Nematodes – Current Status of Mosquito Control Techniques . . . . .	James B. Hoy and James J. Petersen	49
Mosquito Repellents . . . . .	A. A. Khan and H. I. Maibach	50
Aerial Application of Mosquito Adulticides in Irrigated Pastures . . . . .	Mir S. Mulla, Jorge R. Arias, Robert D. Sjogren and Norman B. Akesson	51
Insect Development Inhibitors: Their Potential as Mosquito Control Agents . . . . .	Charles H. Schaefer and William H. Wilder	56
A Comparison of Concentrated Insecticide Solutions and Emulsions for Ultra Low Volume Applications Against Mosquitoes and House Flies . . . . .	E. M. Fussell and S. R. Husted	57
Aerosol Machine Developments . . . . .	Norman B. Akesson	58
Operational Experience with Low Volume Nonthermal Aerosol Generators in The Colusa Mosquito Abatement District . . . . .	Kenneth G. Whitesell	59
Evaluation of Mosquito Adulticides Applied as Nonthermal Aerosols in Irrigated Pastures . . . . .	R. D. Sjogren, M. S. Mulla and J. R. Arias	61
Evaluation of Low Volume Nonthermal Aerosols for Mosquito Control in California . . . . .	Don J. Womeldorf, Ernest E. Lusk, Kenneth R. Townzen and Patricia A. Gillies	67
The Third Era of Mosquito Control in California Has Just Begun . . . . .	Thomas D. Mulhern	75

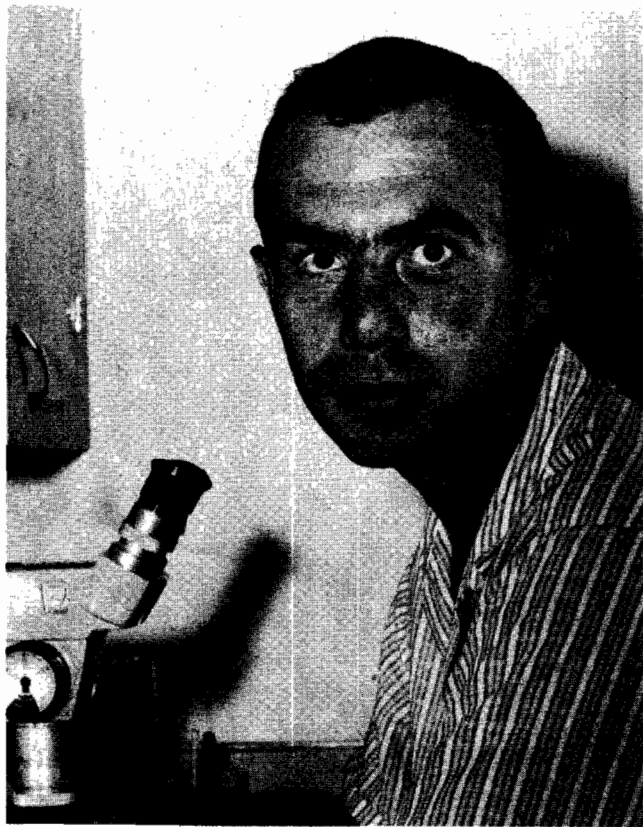
Past, Present and Predictable Future of Aircraft in Mosquito Control . . . . .	Arthur F. Geib	79
Evolving Ecological Awareness as a Basis for Meeting the More Difficult Problems of the Future . . . . .	Gordon F. Smith	80
A Changing Emphasis of Mosquito Control in California: Survey, Planning, Persuasion, Negotiation, Cooperation and Legal Action . . . . .	Jack H. Kimball	84
Advancing Mosquito Control in Difficult Times . . . . .	James W. Bristow	85
A Unifying Framework for Mosquito Control . . . . .	Fred C. Roberts	86
The K-Coefficient: A Measure of Association Between Mosquito Larval Species in Breeding Sources . . . . .	Iver E. Bradley and Jay E. Graham	87
Will Local Governments Be Merged into Regional Governments . . . . .	Marvin C. Kramer	90
Potential Vector Problems Associated with Subsurface Utility Enclosures . . . . .	Charles M. Myers, Ed Colson and Gilbert L. Challet	91
Vertical Drainage for Mosquito Control in 1972 -- A Progress Report . . . . .	Leyburn F. Lewis, Darrell M. Christenson and William M. Rogoff	93
Evolution of a Mobile Communication System . . . . .	E. L. Geveshausen and Gardner C. McFarland	95
Design Aid for Land Grading . . . . .	Harley L. Natvig and Rueben Junkert	96
Successful Source Reduction on Tidal Salt Marshes (Abstract) . . . . .	A. D. Telford and J. D. Rucker	100
Mosquito Source Reduction Helps Cut District Tax Rate . . . . .	T. G. Raley	101
New Mosquito Sources and their Prevention in Urban Areas . . . . .	Gilbert L. Challet	103
Tolerance Thresholds in Mosquito Pest Management . . . . .	R. K. Washino	105
Parous Rates of <i>Anopheles freeborni</i> Populations in Northern California . . . . .	R. J. McKenna, R. K. Washino, C. K. Fukushima and R. P. Meyer	106
Results of Preliminary Mark-Release-Recapture Trials with <i>Aedes nigromaculis</i> . . . . .	G. A. H. McClelland, Robert J. McKenna, Donald J. Jolly and Thomas A. Cahill	107
Susceptibility of Organophosphorus Resistant <i>Culex tarsalis</i> to Experimental Infection with Western Equine Encephalomyelitis Virus . . . . .	J. L. Hardy and R. D. Sjogren	109
Observations on the Hatching of <i>Aedes nigromaculis</i> (Ludlow) Eggs: Seasonal Variation . . . . .	Takeshi Miuri and R. M. Takahasi	111
Biological Control of Aquatic Weeds in the Lower Colorado River Basin. . . . .	E. F. Legner, T. W. Fisher and R. A. Medved	115
Current Progress on <i>Beauveria tenella</i> as a Control Agent for Mosquitoes . . . . .	D. E. Pinnock, R. Garcia and C. M. Cubbin	117
Comparison of Juvenile Hormone Analogue Formulations Against Chironomid Midges under Semi-Field Conditions . . . . .	R. Lee Norland	118
Predation of Mosquitoes and Chironomid Midges in Ponds by <i>Tilapia zillii</i> (Gervais) and <i>T. mossambica</i> (Peters) (Teleostei: Cichlidae) . . . . .	E. F. Legner and R. A. Medved	119
Laboratory Observations and Field Tests with <i>Lagenidium</i> against California Mosquitoes . . . . .	E. M. McCray, Jr., D. J. Womeldorf, R. C. Husbands and D. A. Eliason	123
Harvesting Eggs from Wild Notonectid Populations . . . . .	Richard Garcia	129
Insect Developmental Inhibitors: Effects on Non-Target Aquatic Organisms . . . . .	Takeshi Miura and R. M. Takahashi (Abstract)	130
Use of Dichlorvos Against Organophosphate Resistant Adult Mosquito Populations in Kern County . . . . .	R. D. Sjogren	130
Inoculation of Hydra (Coelenterata) and Predation Effectiveness in Experimental Mosquito ( <i>Culex</i> ) Breeding Habitats . . . . .	Hyo-sok Yu and E. F. Legner	131
Pond Tests with ZR-515: Biological and Chemical Residues . . . . .	C. H. Schaefer, E. F. Dupras, Jr., and W. H. Wilder	137
Laboratory and Experimental and Operational Field Evaluation of Mosquito Larvicides . . . . .	Mir S. Mulla, Husam A. Darwazeh and Robert D. Sjogren	139
Evaluation of Promising Petroleum Oils for the Control of Organophosphorus Resistant <i>Aedes nigromaculis</i> Mosquito Larvae in Irrigated Pastures . . . . .	Husam A. Darwazeh	145

Susceptibility Level of Organophosphorus Resistant <i>Aedes nigromaculis</i> Mosquito Larvae to Petroleum Oils . . . . .	Husam A. Darwazeh	149
Measurement of Propoxur Resistance in Adult <i>Aedes nigromaculis</i> by Time Exposure — A Progress Report . . . . .	E. P. Zboray and K. E. White	153
Effectiveness of a Flit MLO® — Larvicide Oil Formulation in the Field . . . . .	Frank W. Pelsue and Gardner C. McFarland	155

## AUTHOR'S INDEX

Akesson, Norman B. et al. . . . .	51	Junkert, Reuben & Harley L. Natvig	96
Akesson, Norman B. . . . .	58	Kauffman, Eugene E. . . . .	35
Arias, Jorge R. et al. . . . .	51	Khan, A. A. & H. I. Maibach . . . . .	50
Arias, Jorge R. et al. . . . .	61	Kimball, Jack H. . . . .	84
Baier, Dwight C. . . . .	30	Kozlik, Frank M. . . . .	46
Beckus, Robert J. . . . .	1	Kramer, Marvin C. . . . .	90
Bradley, Iver & Jay E. Graham . . . . .	87	Legner, E. F. et al. . . . .	115
Bristow, James W. . . . .	85	Legner, E. F. & R. A. Medved . . . . .	119
Brown, William S. . . . .	39	Legner, E. F. & Hyo-sok Yu . . . . .	131
Burger, Charles H. . . . .	11	Lewis, Leyburn F. et al. . . . .	93
Cahill, Thomas A. et al. . . . .	107	Lusk, Ernest E. et al. . . . .	67
Cappucci, Dario et al. . . . .	13	Maibach, H. I. & A. A. Khan . . . . .	50
Challet, Gilbert L. et al. . . . .	91	McClelland, G. A. H. et al. . . . .	107
Challet, Gilbert . . . . .	103	McCray, E. M., Jr. et al. . . . .	123
Christensen, C. Brunel . . . . .	31	McFarland, Gardner C. . . . .	36
Christensen, Darrell M. . . . .	93	McFarland, Gardner C. & E. L. Geveshausen . . . . .	95
Colson, Ed et al. . . . .	91	McFarland, Gardner C. & Frank W. Pelsue . . . . .	155
Cook, J. Warren . . . . .	32	McKenna, R. J. et al. . . . .	106
Cubbin, C. M. et al. . . . .	117	Medved, R. A. et al. . . . .	115
Darwazeh, Husam A. et al. . . . .	139	Medved, R. A. & E. F. Legner . . . . .	119
Darwazeh, Husam A. . . . .	145	Meyer, R. P. et al. . . . .	106
Darwazeh, Husam A. . . . .	149	Milias, George W. . . . .	2
Day, Boysie E. . . . .	45	Miuri, Takeshi & R. M. Takahashi . . . . .	111
Dietrick, E. J. . . . .	42	Miuri, Takeshi & R. M. Takahashi . . . . .	130
Dupras, E. F., Jr. et al. . . . .	137	Mulhern, Thomas D. . . . .	75
Eliason, D. A. et al. . . . .	123	Mulla, Mir S. et al. . . . .	51
Emmons, Richard W. et al. . . . .	13	Mulla, Mir S. et al. . . . .	61
Fisher, T. W. et al. . . . .	115	Mulla, Mir S. et al. . . . .	139
Fukushima, C. K. et al. . . . .	106	Murray, W. Donald . . . . .	33
Fussell, E. M. & S. R. Husted . . . . .	57	Myers, Charles M. et al. . . . .	91
Garcia, Richard et al. . . . .	117	Natvig, Harley L. & Reuben Junkert	96
Garcia, Richard . . . . .	129	Norland, R. Lee . . . . .	118
Geib, Arthur F. . . . .	79	Pelsue, Frank W. & Gardner C. McFarland . . . . .	155
Geveshausen, E. L. & Gardner C. McFarland . . . . .	95	Peters, Richard F. . . . .	43
Gillies, Patricia A. et al. . . . .	67	Peters, Robert H. . . . .	27
Graham, Jay E. & Iver E. Bradley . . . . .	87	Petersen, James J. & James B. Hoy . . . . .	49
Grodhaus, Gail et al. . . . .	13	Pinnock, D. E. et al. . . . .	117
Hardy, James L. & William C. Reeves . . . . .	7	Raley, Theodore G. . . . .	34
Hardy, James L. & R. D. Sjogren . . . . .	109	Raley, Theodore G. . . . .	101
Hoy, James B. and James J. Petersen . . . . .	49	Reeves, William C. & James L. Hardy . . . . .	7
Husbands, R. C. et al. . . . .	123	Roberts, Fred C. . . . .	86
Husted, S. R. & E. M. Fussell . . . . .	57		
Jolly, Donald J. et al. . . . .	107		

Rogoff, William M. et al. . . . .	93	Telford, A. D. & J. D. Rucker. . . . .	100
Rucker, J. D. & A. D. Telford. . . . .	100	Townzen, Kenneth R. et al. . . . .	67
Schaefer, Charles H. & William H. Wilder . . . . .	56	Washino, R. K. . . . .	105
Schaefer, Charles H. et al. . . . .	137	Washino, R. K. et al. . . . .	106
Silveira, Stephen M. . . . .	37	White, K. E. & E. P. Zboray . . . . .	153
Sjogren, Robert D. et al. . . . .	51	Whitesell, Kenneth G. . . . .	59
Sjogren, Robert D. et al. . . . .	61	Whitten, George R. . . . .	28
Sjogren, Robert D. & J. L. Hardy . . . . .	109	Wilder, William H. & Charles H. Schaefer . . . . .	56
Sjogren, Robert D. . . . .	130	Wilder, William H. et al. . . . .	137
Sjogren, Robert D. et al. . . . .	139	Womeldorf, Don J. et al. . . . .	67
Smith, Gordon F. . . . .	80	Womeldorf, Don J. et al . . . . .	123
Steinmetz, William H. . . . .	38	Work, Telford H. . . . .	3
Swift, John E. . . . .	41	Yu, Hyo-sok & E. F. Legner . . . . .	131
Takahashi, R. M. & Takeshi Miuri . . . . .	111	Zboray, E. P. & K. E. White . . . . .	153
Takahashi, R. M. & Takeshi Miuri . . . . .	130		



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Richard Florin Frolli  
1929 – 1972  
Vector Control Specialist  
California Department of Public Health

- 1972– California Dept. of Public Health: Vector Control Specialist  
1961 – 1971 Kings Mosquito Abatement District: Manager  
1957 – 1961 Corcoran Mosquito Abatement District: Manager  
1954 – 1957 John Muir Junior High School, Corcoran: Educator  
Served as Director of: California Mosquito Control Association  
Member: Hanford Rotary Club  
Leader: 4-H Club  
Active in Boy Scouts, as leader, as executive board member, as chairman, as district representative  
Served as President of Kings Ski Club  
Member: Far West Ski Association Central Council



Neal Gibson  
1920 – 1972  
Manager  
Northwest Mosquito Abatement District

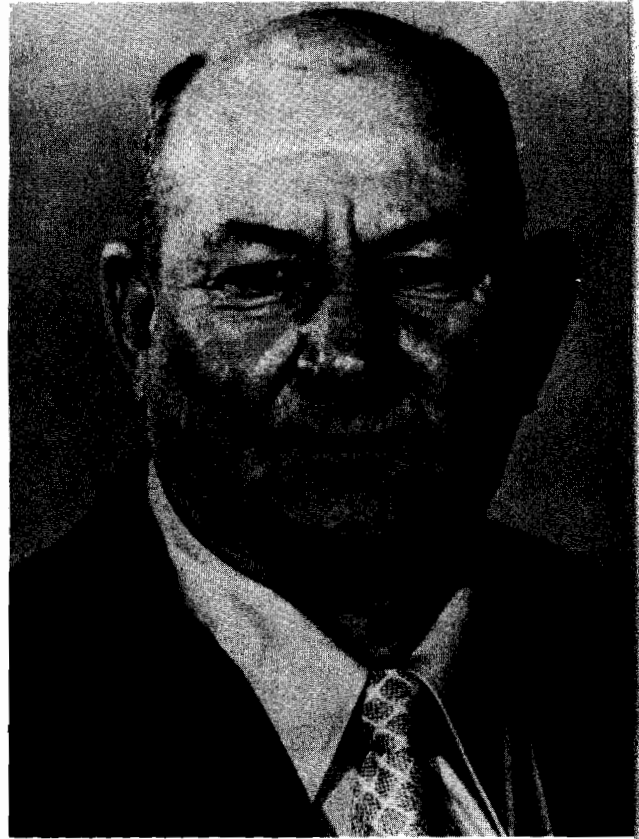
- 1970 – 1972 Northwest Mosquito Abatement District: Manager  
1968 – 1970 Moorpark Mosquito Abatement District: Manager  
1966 – 1968 Glenn County Mosquito Abatement District: Manager  
1964 – 1966 Kings Mosquito Abatement District: Agromonist and Source Reduction Specialist  
1954 – 1964 Wilbur Ellis Company, Fresno: Field Agromonist and Entomologist  
Member: Society of Vector Ecologists  
Member: California Epsilon Chapter of Alpha Zeta Honorary Society  
Past President: Sanger Rotary Club



Howard C. (Pete) Pangburn  
Solano County Mosquito Abatement District  
Manager to 1972

Peter Pangburn, one of the pioneers in mosquito control in California, joined the staff of the Solano County Mosquito Abatement District 42½ years ago. He became manager in 1940. A true sportsman and environmentalist, he was able at an early time to work out methods for integrating mosquito control with marsh wildlife interests, thus winning friends for himself and for his agency. This cleared the path for many cooperative source reduction projects and lasting mutually beneficial relationships with the California Department of Fish and Game and the duck club operators.

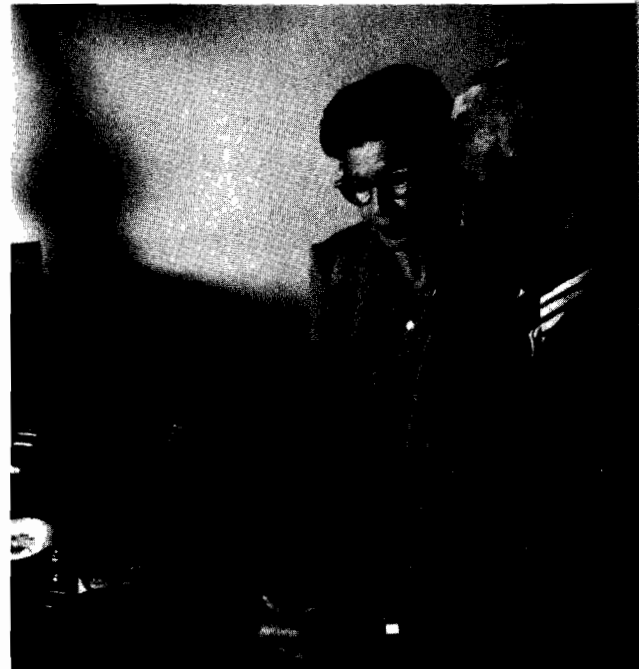
A native Californian whose ancestors were among the historic Donner Party, he has lived most of his life in Solano County, where he is a highly respected community leader. His friends throughout that county and among mosquito control workers throughout the state of California are legion.



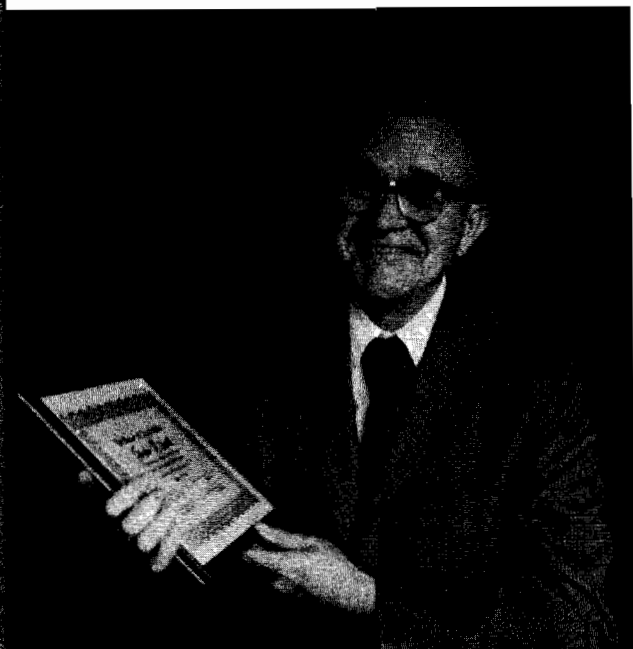
Theodore G. Raley  
Consolidated Mosquito Abatement District  
Manager to 1973

Ted is shown receiving an award of appreciation from the California Mosquito Control Association as his wife Viola shares the moment of honor. Coming in 1947 to the Consolidated Mosquito Abatement District to serve as Manager, Ted has been a dedicated leader not only of that district but also of the California Mosquito Control Association and the American Mosquito Control Association. He was President of the CMCA in 1948 and served on the Board of Directors as Representative of the Southern San Joaquin Valley Region in 1963, 1964, 1970 and 1971. He has been a dynamic member of many committees and has contributed many papers at the CMCA Annual Conferences.

Ted has served as Executive Secretary of the AMCA since 1952 and in this position has aided in the development and coordination of mosquito control throughout this country and the entire world. Prior to his position at the Consolidated Mosquito Abatement District, Ted served one year (1946) as Manager of the Sutter-Yuba Mosquito Abatement District. During World War II he was a technical participant in the Malaria Control in War Areas program.



Gordon F. Smith  
Eastside Mosquito Abatement District  
Manager to 1973



Gordon, affectionately known as "Grumpy" by many of his friends, joined the Kern (then known as Dr. Morris) Mosquito Abatement District in 1946 as the Entomologist. He served as Manager of that district for several years while Arthur F. Geib was on a leave of absence. While at Kern MAD Gordon served on the CMCA Entomology and Insecticide Committees, and was the major force behind the production of a guide for the use of insecticides for mosquito control in California. He was one of the first entomologists in the world to observe and document mosquito resistance to insecticides. Also, he spent many weeks studying the flight patterns of *Aedes nigromaculis* in Kern County.

In 1957 Gordon became Manager of the Eastside MAD. He was President of CMCA in 1959, and served as Representative of the Northern San Joaquin Valley Region in 1963, 1966 and 1971. Although he contributed to many facets of mosquito control in California, Gordon contributed most to the research program. He was on the CMCA Research Committee from 1960 through 1972, and served as Chairman in 1963, 1964, 1965, 1967, 1968 and 1969. Since his retirement, Gordon has escaped the problems and pressures still experienced by MAD managers by going camping, fishing and golfing.

# California Mosquito Control Association

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## THE LEGISLATIVE OUTLOOK RELATING TO MOSQUITO CONTROL

Robert J. Beckus

California Advocates, Inc.

and

California Special Districts Association

925 L Street, Sacramento, California 95814

My relationships with the state legislature usually involve personal contacts with legislators on a man-to-man basis. I represent more than 300 special districts of many types, including mosquito abatement districts, about 15 of which belong to the California Special Districts Association (CSDA). Many more should be members for mutual benefit.

During the 1972 legislative session the California Mosquito Control Association (CMCA) obtained the addition of the word "vector" to the Health and Safety Code. This was not a controversial issue but for a few days private pest control people thought the change might restrict their activities.

Mosquito control agencies are deeply involved in the allocation of taxes as a result of Senate Bill 90, which makes it necessary for local governments to use the referendum process to increase their tax rate above the 1972-73 level. County governments may choose the 1971-72 or the 1972-73 fiscal year as a base. In the near future special districts will probably be given the same option as the county governments.

Mosquito abatement districts are concerned with encephalitis. One of my 17 clients is an association of 1500 breeders of thoroughbred horses. They were gravely concerned in 1971 by the threat of Venezuelan encephalitis, so they are interested in your program and in research to provide technical support for programs to prevent VEE.

The people voted at the last election to put the legislature on two-year instead of one-year sessions. It was said that this would cut the cost of state government, reduce the number of bills, and result in higher quality legislation. I do not believe this is likely to happen. The legislature operates under a committee system and both houses have comparable committees. For example, a bill introduced into the Assembly will go through a policy committee and, if it has financial implications, through the Ways and Means Committee in the Assembly and the Finance Committee in the Senate. To become law a bill has to run this course through both houses and subsequently be signed by the Governor. Under the new rules, which are very similar to

those of the U. S. Congress, a bill introduced in the Assembly can languish there or it can be passed to the Senate at any time up to the last month of the year. The Senate may then take all of the next year to act upon it. If the bill does not get out of the house of origin during the first year, it dies automatically and the author cannot introduce a similar bill during the second year.

The Council on Intergovernmental Relations (CIR) has been mainly a "do-nothing" body, meeting but with little authority. In 1971 it was modified to provide for the addition of three representatives of special districts to be selected by CSDA. The Governor is now giving considerable attention to local government. A steering committee made up of Lt. Governor Reinecke and several agency secretaries will employ a task force of three or four individuals, who in turn will select persons familiar with county, city, and special district governments to serve on the task force. The CIR is going through an educational process and may be expected to give more consideration to the views of the special districts.

Three years ago the Local Agency Formation Commission (LAFCO) law was amended to set up a process whereby special districts could obtain representation. This process was followed in San Diego and Riverside Counties and the program has worked well. However, there are difficulties in San Bernardino, Sacramento, and Ventura Counties, where special districts evidently are not regarded as acceptable bodies of local government and where their representation on LAFCO apparently is not wanted. This year another step is being taken so that where special districts represent a stated portion of the assessed valuation within a county, they will be entitled to representation on each county LAFCO. It is impractical to demand that every LAFCO have special district representation because in some of the 58 counties there are no special districts. However, where special districts are providing essential services, they obviously should be represented in planning for the orderly development of future government organization and services.

## EPA -- ITS IMPACT UPON CALIFORNIA MOSQUITO CONTROL

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The Environmental Protection Agency (EPA) is a fledgling federal agency, a spin-off from the old Water Quality Administration and other environment-related activities. One part is involved in the research and technical aspects of environmental problems and employs a large research staff. The other portion is involved in the role of policemen of the environment. The Congress of the United States passed some very strong legislation during the past three sessions. Individual congressmen should not be blamed for some of the language that went into the Act, but the technicians who drafted the legislation managed to get many kinds of little angles and quirks into it. EPA must first try to interpret what Congress really meant, then how this interpretation can be carried into effect without literally attempting to shut down the world.

Mosquito control agencies will occasionally come into contact with EPA, particularly in relation to changes in the law on insecticides and herbicides. The new 1972 act completely revised the Federal Insecticide Fungicide and Rodenticide Act (FIFRA), which has been the basic authority for pesticide regulations since 1947. Under the new act, two use categories for pesticides are set up -- general and restricted. Restricted use pesticides can be used only by certified applicators. These are products which could cause substantial adverse effects on the environment and which require more knowledge and care on the part of the individual applying them.

Minimum standards for the certification of applicators will be set by EPA and will be separate for private and commercial applicators. Certification will be accomplished by states whose programs are approved by EPA. Collection of information on state programs for certification of applicators is now in progress. Present schedules call for the com-

pletion of preliminary certification standards within seven months. The act requires that final certification standards be published within one year. This portion of the new act should reduce misuse of pesticides and eliminate or reduce pesticide accidents; it will correct the "all or nothing at all" approach now in effect. Rather than being forced to cancel or suspend a product outright, the only recourse under FIFRA, EPA now may be able to allow registration of a much needed product under the restricted category. Federal authority will apply throughout the United States, not just to the movement of products across state lines. The act will require the registration of all pesticide-producing establishments and regular submission by them of production and sales volume information.

Pesticides are a product of today's age of technology which has often progressed faster than has the knowledge of how to use these products wisely. It is imperative that the future uses of new technology do not result in irreparable damage to the environment or to human beings.

There is now demonstrable evidence of cooperation among the various levels of government. EPA prefers this approach because it does not want to do things which the states, the districts or other local agencies can do better for themselves. In California, with the cooperation of the California Department of Food and Agriculture and of the federal authorities that are involved, everyone will try to do his job with a minimum of harrassment of the people, yet providing the level of protection that is necessary. EPA urges that those who have problems present the facts and keep the lines of communication open.

# THE IMPENDING THREAT OF VEE TO CALIFORNIA BASED ON THE MEXICAN EXPERIENCE OF 1972<sup>1</sup>

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On a foggy, overcast day in January of 1972, this Association met in Stockton to hear reports of data on mosquito transmitted viruses in California during 1971, and a speculative discussion about the significance of the Texas experience with Venezuelan equine encephalitis. Concerns focussed on two main questions: 1) Can it happen in California? 2) Is the state equine and human population safe as a result of vaccinating 485,000 California equines with live attenuated TC83 VEE vaccine in August and September 1971?

The reports by Reeves, Emmons, and Work established that the rich variety of ecological situations ranging from Tehama County of northern Sacramento Valley, and Inyo County east of the Sierras to Imperial County on the Border of Mexico annually support mosquito transmission of western equine (WEE), St. Louis (SLE) or California encephalitis (CE) viruses. It was also reported that the ubiquitous use of any mosquito by VEE in an epizootic-epidemic situation could well transform the summer months into a vector-borne donnybrook of Venezuelan equine encephalitis, should this virus reach California via the west coast of Mexico, where its existence was not yet known to be established. At that time there were doubts as to the possibility of VEE ever reaching the Pacific Coast. These doubts were based on two assumptions: 1) That since the initial invasion into Chiapas, Mexico, from Guatemala in 1969, VEE had not extended along the Pacific Coast; and 2) that the Sierra Madre Occidental was a natural barrier limiting the widespread activity of 1969, 1970 and 1971 along the Mexican Gulf Coast and on the Central plateau from ever reaching the west coast.

Now, at the end of January 1973 we meet again to consider the same questions, but with considerably more knowledge and experience derived from the fact that VEE did reach the Pacific Coast of Mexico in 1972 and that in the period June through September, it progressed northwestward a distance of about 700 miles leaving in its wake thousands of dead horses and thousands of human cases — with some deaths. That this relentless progression occurred through a narrow coastal plain corridor, where tens of thousands of equines had been vaccinated, presents a question as to whether equine vaccination really is a barrier in which people can have confidence for blocking movement and dissemination of this mosquito borne virus.

Like so many advances in science, this enterprise resulted from a fortuitous but timely integration of a variety of interests and resources. Dr. Luis Fernandez-Zorilla, Veteri-

nary Jefe of the Equine Encephalitis Program of the Department of Agriculture in Mexico, had taken his Master of Public Health degree in June 1971 from UCLA where he had special field and laboratory training in arboviruses of the Imperial Valley. A National Institutes of Health Training Grant had supported these investigations with a number of entomological and virological research trainees, some of whom were in place and ready to undertake investigations in Mexico. The long term scientific contributions initiated and sustained by the University of California — particularly those of Reeves in San Francisco and Berkeley — had housed and supported development of university resources for research on this abiding public health problem in California, most recently by the provision of laboratory facilities at UCLA. The U. S. Department of Agriculture Veterinary Services Emergency Programs had committed funds, through Professor George Crenshaw of UC Davis to carry out surveillance for VEE in California. This was initiated in January 1972 by a joint UC Davis — UCLA Program in Palo Verde, Yuma and Imperial Valleys by placement of sentinel equines and periodic mosquito collections for serological and virological studies. (Bown and Work, 1973).

A research training operation in collaboration with our Mexican colleagues northward from Mazatlan had been scheduled for June after completion of the academic year. So all of the technical, scientific, professional and material resources were in hand in June. What remained to pull them together was the precipitating report that Venezuelan equine encephalitis had erupted on the Pacific Coast of Mexico.

A week before the scheduled departure of our field research training group for Mazatlan, Dr. Fernandez telephoned to report that on June 16, equine deaths had been reported from three coastal municipalities in Nayarit; Tuxpan, Santiago and San Blas. He inquired as to whether we would be interested in extending our itinerary to Nayarit to undertake collaborative studies with our Mexican veterinary colleagues. This changed the level of commitment in regard to preparation and time. With TC-83 vaccine obtained from the National Center for Disease Control, we vaccinated thirty-four staff and students on June 29, after which Alfred Sierra and Peter Warner, the veterinary student team from UC Davis, and our students, departed by road with vehicles and equipment for Tepic. By air through Puerta Vallarta I joined Dr. Fernandez in Nayarit to assess the developing epizootic as he implemented a statewide vaccination of about 30,000 equines.

This commenced collections and studies that were to occupy us in the field from Nayarit, through Sinaloa, and the length of Sonora, to the Arizona and California Border in the next three months, as the disease erupted in various facets northwestward in June from Rosamorada, in Nayarit,

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Esquinapa in Sinaloa; in July, the Fuerte Valley near Los Mochis; in August, Huatabampo and Navjoa of southern Sonora and in September Soyopa and La Misa. This was accomplished by rotation of student and staff personnel; those spending the longest periods being entomologists Dr. David Bown and Dr. Bruce Knudsen and Dr. Victor Schroeder of the Animal Health Division of the Mexican Department of Agriculture.

The effort was focussed on three main problems. 1) Was the Mexican equine vaccination program an effective barrier to movement of the virus? 2) What other vertebrate-mosquito mechanisms might be involved in movement of the virus? 3) After introduction of VEE virus into a rural area what other peridomestic vertebrates provided an adequate viremia for infection of vector mosquitoes that in turn transmit VEE virus to human inhabitants of a village?

The first equine isolation of VEE virus at UCLA was from serum collected on July 5 from a paralyzed burro at Teponautla in Rosamorada Municipality of Nayarit. It confirmed the northward movement of the epizootic, augmented by observation of sick and dead horses alongside Highway 15, the western route of the Pan American Highway, only three miles from Rosamorada.

Reports of equine disease from El Fuerte in northern Sinaloa led to investigations which found sick and dead animals in the mountainous western slopes of Sierra Madre Occidental. A serum from a moribund burro collected July 18 in a newly cleared corn and bean field above Chinobampo had HAI and NT antibodies to VEE virus. This epizootic was in contrast to those in tropical Nayarit which began in the coastal lowlands and moved up river drainages to higher elevations.

The Chinobampo outbreak occurred in the highlands of northern Sinaloa, the only apparent connection being via the transcontinental railway from Ojinaga (across the Rio Grande River from Presidio, Texas, where the last U. S. equine isolate of VEE was made on November 7, 1971) via Chihuahua to Los Mochis and the Pacific Port of Topolobampo. This epizootic moved down the Fuerte River to the delta, adjacent to the southern border of Sonora where equine deaths were reported in early August.

An extensive epizootic epidemic, involving more than six thousand human cases with at least six reported deaths was described from nearby Navajoa area in the last week of August. Dr. Knudsen's collecting effort and probable area of exposure was in this area where there was an afflicted fishing community that had only one resident horse.

Dr. Bruce Knudsen, one of our postdoctoral trainees, was between 25th and 30th in order of inoculation of the 34 vaccinations on June 29th. Yet at 2:00 a.m. on August 24 about 30 hours after collecting mosquitoes in the Navajoa area of Southern Sonora where the new epizootic-epidemic of VEE was developing, Dr. Knudsen had sudden onset of fever, headache and prostration. On arrival at UCLA in the evening of August 25 a blood specimen was collected. Inoculation into suckling mice yielded a strain of VEE virus.

Serial sera collected periodically during the previous eighteen months indicated what was probably a field infection with western equine encephalitis virus in late June 1971 during an eruption of WEE in Imperial Valley. He had

sustained a short, self limited illness at that time. Serological testing of pre- and post vaccination sera of twenty-four of the thirty-four vaccinees showed conversion to VEE antibody. Pre-existing WEE antibody, resulting from natural infection a year before, was apparently responsible for the refractory response to arbovirus group A live virus VEE vaccine in Dr. Knudsen.

Our studies obtained further information on this point. Early in July, when the effectiveness of the Mexican VEE vaccine was in question, we outlined a test of sixty horses in the vicinity of Hermosillo where a vaccination campaign was underway. Drs. Hidalgo and Valdez Mungia of the Hermosillo Veterinary Laboratory collected sera from sixty marked equines just prior to vaccination. They were re-bled approximately one month post vaccination. The paired sera were tested for hemagglutination inhibiting (HAI) antibodies against VEE, WEE, Sindbis and St. Louis antigens. Neutralization tests were run in tissue culture.

Four of fifteen equines with demonstrable pre-existing WEE virus antibody failed to respond to the VEE vaccine with rise in antibody titer compared to only one of thirty eight animals without pre-existing WEE antibody. Because the Mexicans do not routinely vaccinate for WEE it was assumed that the pre-existing WEE antibody resulted from natural WEE infections in the Hermosillo area.

Not only do these findings illuminate a problem to be encountered in vaccination programs in Mexico, but they cast doubt on how many equines vaccinated for VEE in southwestern United States in 1971 were protected because there are significant numbers of equines that have measurable titer of WEE antibody due either to routine vaccination or natural infections. Study of this problem continues.

It should be observed here that epizootic-epidemic activity from June to late August was observed in the narrow coastal corridor from the Santiago-San Blas Municipalities of Nayarit in June, northward through Rosamorada, Esquinapa, Mazatlan and El Fuerte in Sinaloa in July to Navajoa in August. Many rivers run from the Sierras to the sea, from east to west, transecting this corridor. The Navajoa area, where the sizeable outbreak occurred in late August, and Ciudad Obregon encompass the delta of the Yaqui River which flows south from Arizona and western New Mexico. Geographically this delta is the southern limit of the Sonoran Desert which fans out to the north and northwest into an irrigated chain of agricultural developments that link with the Colorado Delta, Yuma Valley and the Imperial Valley, where we know, from previously described investigations initiated by UCLA in 1967, there is evidence of almost continuous mosquito transmission of a variety of arboviruses.

Although the destination for the September investigations was the water bird inhabited estuaries of coastal Sonora, the increasing occurrence of equine disease east and south of Hermosillo called for case investigations for collection of virus containing specimens to document temporal dispersion of the virus. This took our field teams east of Hermosillo to the town of Soyopa, on the north-south course of the Yaqui River a natural avenue northward for virus movement from the Navajoa epizootic of late August. Among the many affected animals in this area, a blood specimen was collected from an equine forty-five minutes after

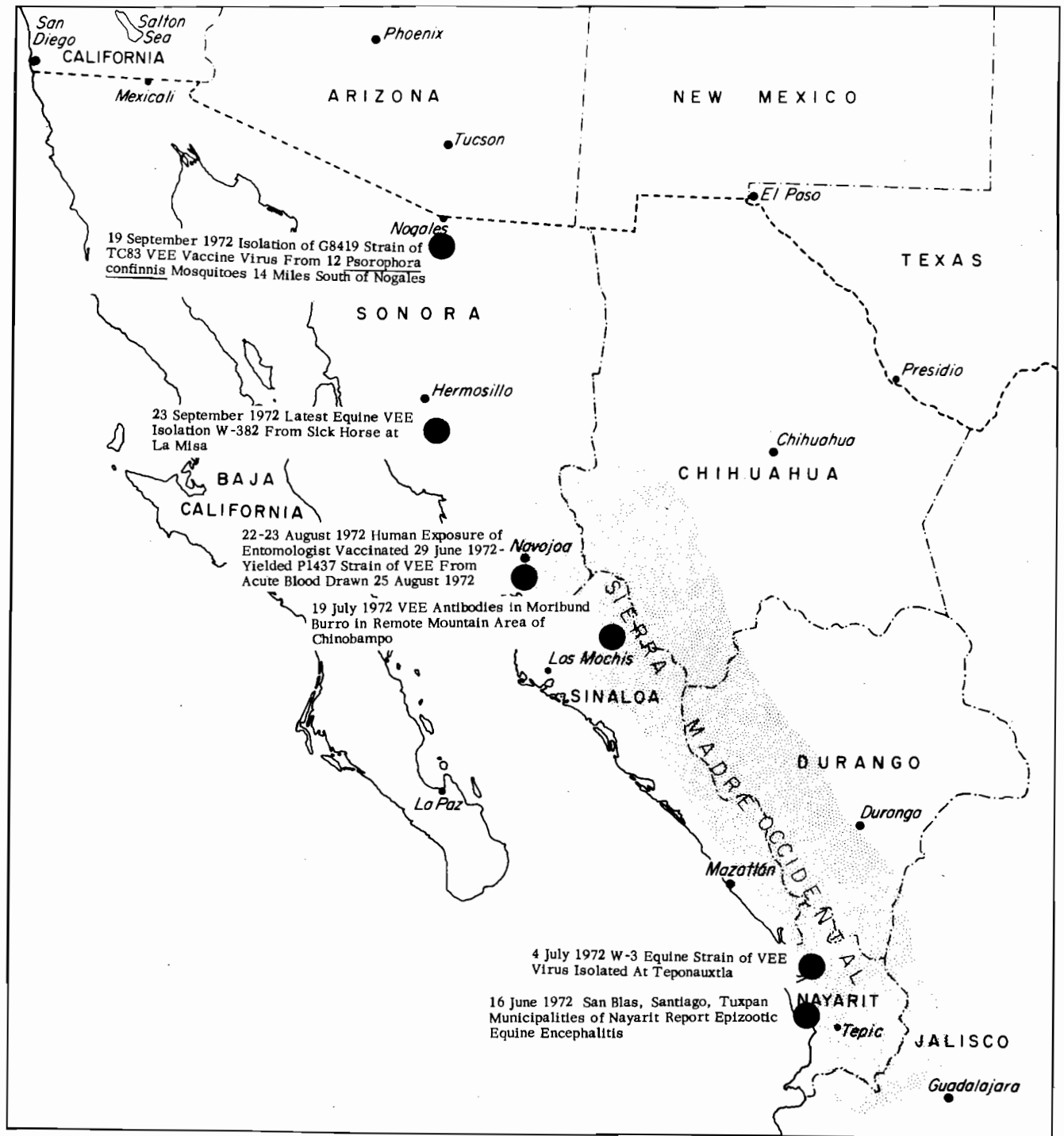


Fig. 1.—

death. From this serum an agent (W-311) that kills suckling mice in less than 24 hours was isolated. It is not VEE virus. Its significance in these studies is that there may well be another agent, fatal for equines, producing disease that has been reported as VEE. It would also explain why apparently VEE immunized equines subsequently died in an equine epizootic.

The probable field infection of one of our own personnel, followed by the extensive late August epizootic epidemic in Southern Sonora, called for a shift in field activity to this border state adjacent to the susceptible areas of southwestern United States. Surveillance for equine disease and efforts to vaccinate equines in Sonora were intensified by the Mexican Department of Agriculture in July. In Septem-

ber equine cases were reported just south of Nogales. A vaccination campaign carried out in a 70 kilometer radius from Nogales on the first and second of September vaccinated 1,633 equines. A ten trap mosquito collection on the night of 18-19 September just 14 miles south of Nogales yielded few mosquitoes. But a pool, G8419, of 12 *Psorophora confinnis* produced death in inoculated suckling mice in less than 48 hours. HA testing of a borate saline antigen of this agent indicated that it was a strain of VEE. The agent was reisolated from the original mosquito suspension.

One second passage mouse brain harvest each was dispatched to the USPHS Center for Disease Control in Atlanta and the USDA Animal Disease Research Laboratory in Denver for virulence testing to determine whether the virus was a wild or vaccine strain. Parallel testing was carried out at UCLA. Failure to kill guinea pigs by intraperitoneal inoculation indicated that the mosquito isolate was a vaccine type. This is the second isolate of a vaccine strain from *Psorophora confinnis* mosquitoes; the first being in western Louisiana in 1971 following extensive equine vaccination during the Texas epizootic. It appears that this G8419 strain is a biological replicate from *Psorophora confinnis* mosquitoes. VEE vaccination was carried out on September first and second in this particular area. Vaccine virus viremia would occur two to five days later or prior to September 8. Extrinsic incubation, allowing for loss of ingested virus in the stomach in 48 hours, would have occurred by September 18, the night the mosquito traps were set which caught the *Psorophora confinnis* mosquitoes from which the isolation was accomplished.

The significance of this isolation consists of 1) Demonstration that systematic mosquito collection related to epizootics is a sensitive indicator of virus infection of mosquitoes, 2) that some vaccinated equines circulate TC-83 virus in high enough titer to infect mosquitoes and 3) that mosquitoes may transmit the attenuated strain of VEE virus in nature. The questions raised pose further problems to be investigated.

**CONCLUSIONS.**—Only the highlights of this past year's investigations and findings have been touched upon here to illuminate some of the problems in predicting whether there is an impending threat of VEE to California in 1973. On the one hand we have extensive evidence of the appearance and northwestward progression of VEE on the Pacific Coast of Mexico in 1972, with subsidence in Central Sonora late in September. On the other hand intensive search for VEE in Texas where it subsided at a similar latitude in 1971 failed to produce any evidence of a recrudescence in 1972.

Although our investigations demonstrated the availability of virus transmission by infected mosquitoes or viremic individuals in vehicular traffic along Highway 15, only a steady progression northward, seven hundred miles from June through September appears to have occurred; scattered epizootics having been observed some distance from this main artery of traffic.

Virological examination of mosquito collections yielded a vaccine strain fourteen miles south of the Nogales, Arizona, border. This demonstrates the value of VEE virus surveillance by mosquito collections but does not provide clear implications of isolating vaccine virus from *Psorophora confinnis* vectors.

Similar mosquito collections and isolations in selected sites of the arid zone of Southern California, adjacent to the Mexican Border, have yielded numerous strains of WEE and SLE viruses, but no VEE. This again confirms that the mosquito ecology of California will support VEE virus transmission should this virus reach our territory.

Perhaps of greatest importance is the cumulative evidence that pre-existing western equine encephalitis virus antibody produces a refractory response in many animals to TC-83 VEE vaccine. Recognizing that many California equines possessed WEE antibody before this VEE vaccination campaign of 1971 leaves us uncertain as to how many California equines are protected and how many susceptibles there are that would serve as virus circulators of VEE virus for infection of mosquitoes.

With these uncertainties it is essential to return to the question of vector control should VEE resume its northwestward progression into Arizona and California. The apparent indiscriminate use of any mosquito species by VEE virus in an epizootic-epidemic challenges mosquito control resources with more than vector specific measures which have been the mainstay of mosquito abatement in the past.

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# EMERGING CONCEPTS OF FACTORS THAT LIMIT THE COMPETENCE OF *CULEX TARSALIS* TO VECTOR ENCEPHALITIS VIRUSES<sup>1</sup>

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Most of the participants of this conference will remember that an epidemic of encephalitis was anticipated in the Central Valley of California during the summer of 1969. The principal ecologic indicators (rain, snow surplus and flooding) that are used to predict epidemics of western equine encephalomyelitis (WEE) and St. Louis encephalitis (SLE) were apparent during the preceding winter and spring months (Reeves 1967, 1970). An emergency program was initiated in the spring and continued through the summer to increase mosquito control efforts, to assure surveillance for disease in man and horses and to measure viral infection rates in the primary vector mosquito, *Culex tarsalis*. An encephalitis epidemic did not develop. In fact, WEE viral activity was at the lowest level ever recorded in California up to that time and SLE viral activity was found only in the Sacramento Valley (Lyness 1970, Nelson 1970, Reeves 1970, Sudia et al. 1971).

The most puzzling aspect of the 1969 encephalitis season was that WEE and not SLE virus virtually disappeared from the Central Valley and this situation recurred in 1970. The dramatic decrease in WEE viral activity was completely unpredicted because previous studies in Kern County had indicated that higher population levels of *C. tarsalis* were required to maintain the transmission cycle of SLE virus than were required to maintain the transmission cycle of WEE virus (Reeves 1971). Furthermore, retrospective analysis of the 1969 encephalitis season revealed that *C. tarsalis* population levels were sufficiently high at a number of sites in Kern County to support the transmission cycle of WEE virus, even though it disappeared (Lyness 1970). Thus, it became apparent that factors other than population threshold levels could influence the competence of *C. tarsalis* to vector WEE virus. Studies to elucidate these factors and to determine their impact on vector competence were initiated during the past two years.

**FACTORS AFFECTING VECTOR COMPETENCE.**—By vector competence we mean the ability of a mosquito to become infected with a virus and subsequently to transmit the virus to a vertebrate host after an appropriate period of extrinsic incubation. Mosquitoes that are capable of being infected with low concentrations of virus presumably would be more competent vectors than mosquitoes that required high concentrations of virus to become infected. It has long been recognized that *C. tarsalis* was a more competent vector of WEE virus than other mosquito species that prevail in California. We are now suggesting that the vector compe-

tence of different populations of *C. tarsalis* might vary and if so the present problems in controlling this mosquito make it urgent that we learn what controls competence. Factors that could potentially influence the vector competence of *C. tarsalis* and that we are currently studying are shown in Figure 1.

One factor that correlated with decreased WEE viral activity in California during 1969 and 1970 was the emergence of a population of *C. tarsalis* that was highly resistant to the available organophosphorus (OP) insecticides (Georghiou et al. 1969, Womeldorf et al. 1972). We initiated a study to determine if OP resistant *C. tarsalis* were less competent vectors of WEE virus than were OP susceptible populations.

A second variable to be studied was autogeny. This term refers to the ability of a female mosquito to lay her first batch of eggs without taking a blood meal and is a common event with *C. tarsalis* (Bellamy and Kardos 1958, Chao 1958). Lyness (1970) found that autogeny rates in *C. tarsalis* were high during the summer of 1969 in Kern County. The significance of this latter observation was unknown because there was little baseline information available on autogeny rates in *C. tarsalis* populations during previous years. Nonetheless, since high autogeny rates in a vector population could dampen the transmission cycle of an encephalitis virus by delaying the taking of an infectious blood meal, we are attempting to evaluate the impact of autogeny on vector competence.

Quite by accident, Dr. Edward S. Sylvester and Miss Jean Richardson in the Department of Entomology and Parasitology at the University of California at Berkeley found that our colony of *C. tarsalis* in Bakersfield was infected with its own viruses and these were not encephalitis viruses. Virus must be transmitted through the egg because it is found in larvae, pupae and adult males and females. Preliminary studies have suggested that this indigenous mosquito virus might cause degeneration of the salivary glands. If field populations of *C. tarsalis* are infected with a similar virus, then this could potentially result in decreased transmission rates of encephalitis viruses. It is also possible that the "mosquito viruses" might block infection of mosquitoes with a second virus, such as WEE, when it is ingested.

It is conceivable that the susceptibility of *C. tarsalis* to infection with encephalitis viruses is genetically determined. This concept is not revolutionary because genetic susceptibility has been demonstrated for insect vectors of some plant viruses (Storey 1932, Black 1943, Nagaraj and Black 1962). At present we are just beginning studies to examine this possibility.

Environmental conditions, such as temperature and humidity, undoubtedly affect vector competence but the exact extent has not been determined. In addition to its effect on mosquito development and longevity, temperature affects

<sup>1</sup>This research was supported in part by Research Grant AI 03028 from the National Institute of Allergy and Infectious Diseases, and the General Research Support Grant 5-S01-RR-05441 from the National Institutes of Health, U. S. Department of Health, Education and Welfare.

the rate of multiplication of virus in the mosquito and the length of time required to complete viral incubation that ends in an ability to transmit by bite. Hess et al. (1963) reported that annual transmission rates of WEE and SLE viruses in sentinel chickens were closely related to spring temperatures. Highest transmission rates of WEE virus occurred during years when spring temperatures were unusually low whereas highest transmission rates of SLE virus were recorded in years when spring temperatures were unusually warm. The reason for this is not clear. Reeves and Hammon (1962) reported the essential relationship between 80°F and 85°F average daily temperatures for effective transmission of WEE and SLE viruses respectively in the summer.

#### APPROACHES TO VECTOR COMPETENCE STUDIES.

—Both field and laboratory strains of *C. tarsalis* are being evaluated for resistance to five OP insecticides, for autogeny rates, for infection with indigenous mosquito viruses, and for susceptibility to infection with WEE, SLE and Turlock viruses by intrathoracic inoculation and by feeding on gauze pledgets soaked with a suspension of virus in defibrinated rabbit blood plus 1.5 percent sucrose to entice the mosquitoes to feed. Sampling of field populations has depended greatly on collaboration with managers and staffs of numerous mosquito abatement districts in the Sacramento, San Joaquin, Imperial, and Owens valleys. Wherever possible, miscellaneous sampling of field populations was done on the basis of different geographic locations, anticipated levels of resistance to OP insecticides, and, of course, availability of large numbers of *C. tarsalis* larvae and pupae. During 1972 *C. tarsalis* were sampled periodically throughout the summer from two sites in Kern County to look for seasonal variation in vector competence.

**PRELIMINARY RESULTS.**—Our initial studies indicated that four colonized strains and four field strains of *C. tarsalis* were equally susceptible to infection with WEE virus when the virus was inoculated intrathoracically. However, when mosquitoes were allowed to ingest virus from pledgets, the different strains of *C. tarsalis* varied greatly in their susceptibility to viral infection. These observations indicated that variations in susceptibility probably occur in the gut rather than throughout the mosquito.

Variations in the susceptibility of different populations to infection when fed virus is illustrated by comparing the susceptibility of two field strains of *C. tarsalis* collected in Kern County during late August 1971. One sample was collected from a drainage sump at the lower end of a pasture immediately south of Bakersfield that utilized sewage effluent for irrigation. The other sample was collected from a pasture along Poso Creek that utilized effluent from oil wells for irrigation. The percent of mosquitoes infected 14 days after feeding on varying concentrations of virus was determined for each population. When the amount of virus necessary to infect 50 percent of the mosquitoes by pledget feeding was compared, the "Poso Creek" sample was approximately 10,000 fold more resistant to infection with WEE virus than was the "Sewer Farm" sample. It is of interest that WEE viral activity has rarely been detectable in the Poso Creek area even though this area was characterized as having large vector and avian populations. Perhaps this lack of WEE viral activity was related to the fact that *C. tarsalis* from this area are relatively resistant to infection with WEE virus.

Another interesting observation was that about 75 percent of the mosquitoes from the "Sewer Farm" became infected when they fed on 10 to 1,000 infectious units of virus whereas the remaining 25 percent were resistant to infection even when the concentration of virus was increased by 1,000 fold. Similar results were obtained with a strain of colonized *C. tarsalis*. These observations indicated that mosquito populations could be made up of virus susceptible and virus resistant components, which could be genetically determined. If this is indeed true, then perhaps certain environmental factors, such as temperature, could select for the virus susceptible or resistant component in a population.

We were unable to show any correlation between susceptibility to infection with WEE virus and resistance to OP insecticides in *C. tarsalis* from either field or laboratory sources. In fact, the data obtained with field populations from Kern County suggested that OP resistant populations of *C. tarsalis* were more susceptible to infection with WEE virus. However, none of the laboratory or field strains of *C. tarsalis* had been stressed or pressured recently with OP insecticides. In preliminary studies, it was found that fenitrothion stressed laboratory strains of *C. tarsalis* and *Culex quinquefasciatus* were more resistant to infection with WEE virus than were their nonstressed counterparts. Further studies are needed to confirm these results and to determine if similar results can be obtained under field conditions.

Autogeny rates in field populations of *C. tarsalis* collected in California during 1972 are shown in Table 1. Autogeny rates in the "Sewer Farm" population were high throughout the summer whereas those from "Poso Creek" were somewhat lower. Of interest was that less than one percent of a sample collected from a gravel pit in Los Angeles County in July were autogenous. The significance of these results is not clear at this time because autogeny is greatly influenced by larval nutriment and probably is genetically controlled (Spielman 1971). We have not found any correlation between virus susceptibility and autogeny rates in field populations. A preliminary analysis of data from 1971 would suggest, however, that OP resistant populations of *C. tarsalis* tend to be more autogenous. This observation requires further study before a definite conclusion can be drawn.

**IMMEDIATE AND FUTURE CONCERNS ABOUT ENCEPHALITIS IN CALIFORNIA.**—We would be derelict in our obligations to the participants of this conference if we did not mention the pending threat of an epidemic of encephalitis in the Central Valley of California during the summer of 1973. There was a resurgence of WEE and SLE viral activity in the Central Valley during 1972, especially in the San Joaquin Valley (Emmons 1973). This was indicated by the increased number of human cases of WEE and SLE, by numerous isolations of encephalitis viruses from *C. tarsalis* and by the demonstration for the first time since 1968 of immunologic conversions to WEE and SLE viruses in sentinel chickens maintained in Kern County. As of January 1973, we appear to be headed for a record year of rainfall and snowpack which will increase the water availability for mosquito breeding during the summer of 1973. We continue to be faced with an inability to effectively control population levels of *C. tarsalis* due to their resistance to OP insecticides and to the lack of alternate control methods (Schaefer

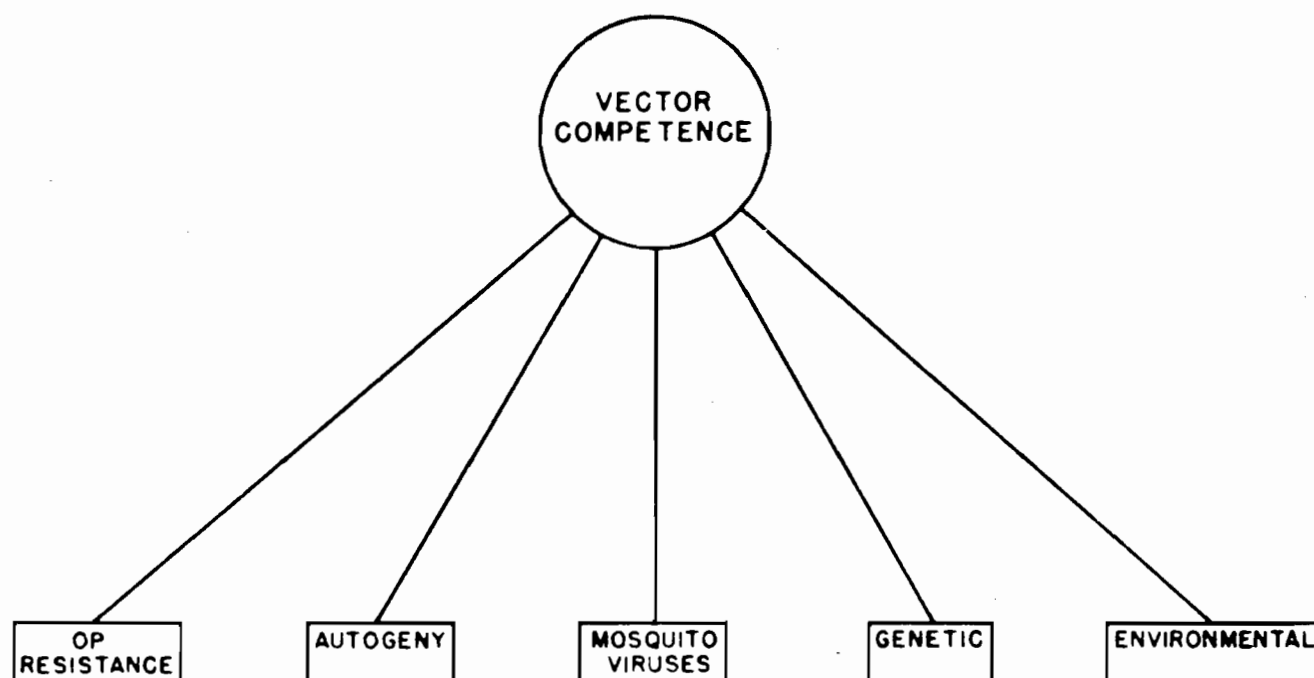


FIGURE 1 — FACTORS INFLUENCING VECTOR COMPETENCE OF CULEX TARSALIS

Table 1.—Autogeny rates in wild strains of *Culex tarsalis* collected in California, 1972.

Mosquito Source	Autogeny rate by month of collection				
	May	June	July	August	September
Kern County					
Sewer Farm	70	79	87	66	90
Poso Creek		44	54	47	67
Meadow Gold				63	
Panama Lane				33	
Richfield				64	
Glenn County					
Road P			32		
Los Angeles County					
Flintkole Gravel			61		
Tulare County					
Correia Ranch			81		
Stanislaus County					
Freitas Pasture			22		
Butte County					
Boeger's Pasture				22	
Yuba County					
Bingham's Ditch				24	
Shintaffer's Hog Farm				87	
North Beale Road				78	

1972). In addition, the immune status of the human population at risk is unknown but is probably low because there has not been a major epidemic or high level of endemic transmission of encephalitis viruses in California since 1952 and 1958.

As to the future control of encephalitis in California, we will need to revise our thinking as more information becomes available on the factors and, hopefully, mechanisms that regulate vector competence. If poor vectors can be made the predominant population, then we may have to live with numbers of *C. tarsalis* just below the pest threshold level--and that represents fairly large populations since *C. tarsalis* is not particularly a pest mosquito. How this can be accomplished is uncertain, but perhaps we will have to introduce genetically poor vectors into the population or even perhaps continue to stress OP resistant mosquitoes with more OP insecticides.

#### Acknowledgment

In closing, we would like to acknowledge some of the people in this audience who have contributed greatly to our program, without whose collaboration our research program would have been greatly hampered. These include, among others: Mr. Melvin L. Oldham, Tehama MAD; Dr. William E. Hazeltine, Butte County MAD; Mr. Eugene E. Kauffman, Sutter-Yuba MAD; Mr. Stephen M. Silveira, Turlock MAD; Dr. W. Donald Murray, Delta MAD; Mr. Arthur F. Geib and Dr. Robert D. Sjogren, Kern MAD; Mr. Sidney H. Ryall, West Side MAD; Mr. George S. Stains, Coachella Valley MAD; Mr. Ben F. Keeney, Inyo Co. Health Department; Dr. Mir S. Mulla, U. C. Riverside; and Dr. Charles H. Schaefer, U. C. Mosquito Control Research Laboratory.

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## THE VETERINARY OUTLOOK CONCERNING ENCEPHALITIS PREVENTION

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The Western Livestock Journal of November 20, 1972, reported that Earl Butz, Secretary of Agriculture, declared an end to the Venezuelan equine encephalitis (VEE) emergency which had been in effect since July 1, 1971, when the disease was first reported in southern Texas. The quarantine was also lifted on Texas equines, the last case having been recorded on November 7, 1971 in Starr County, Texas. VEE had been a serious threat to the horse population of the United States, a population predicted to double to 14,000,000 by 1980.

Prior to the 1971 VEE epizootic in Mexico and Texas, the interest of many practicing veterinarians in equine encephalitis vaccinations lay somewhere between complacency and apathy. Very few cases of Western equine encephalitis (WEE) have been diagnosed in California in recent years, and vaccination was a low revenue procedure. VEE changed this and three million horses were vaccinated in a relatively short time in 19 states. Quarantines were enforced in six states, Texas, Louisiana, Arkansas, Oklahoma, Mississippi, and New Mexico. There was a large supply of TC 83 vaccine and the technology to manufacture more as the need arose. In 1969 over one half million doses of TC 83 vaccine were used in Guatemala, El Salvador, Nicaragua and Honduras. This constituted a very extensive field test.

Controversy over this vaccine did arise. The Grayson Foundation for Equine Research and the University of Kentucky issued reports that four of eleven vaccinated horses showed brain damage on post mortem examination. This report later was amended to state that only one showed severe brain hemorrhage which may or may not have been caused by the vaccine. Thoroughbred owners, aware of the original Grayson Foundation report were reluctant to have their horses vaccinated and some difficulties did arise over this sketchy report and the way in which it was released to the popular horse press.

The U. S. Department of Agriculture vaccinated 45 horses and sacrificed them at intervals between 10 and 49 days post vaccination. No brain damage or spinal cord lesions were found. The Jen-Sal Laboratory, which manufactured the vaccine, vaccinated 20 horses with a higher than normal dose and kept 10 for controls. At autopsy, the brains and spinal cords showed no evidence of vaccine-produced lesions. Jen-Sal used field strength vaccines and found no lesions in 8 horses sacrificed 100 days after vaccination. A third very important study by Jen-Sal tested 702 mares in various stages of pregnancy. Thirty-one were found barren by serological tests, 653 delivered foals that were considered normal at birth, and 18 aborted after vaccination. The abortions occurred 23 to 198 days post vaccination. Four of these were diagnosed as rhinopneumonitis virus, seven mares aborted twins, four had placentitis, one

abortion was a septic foal, one mare was kicked, and one abortion was undiagnosed. The laboratory concluded "there was no evidence that the vaccination induced abortion, fetal abnormalities, or premature foals."

The American Association of Equine Practitioners indicated at its conference held on June 7 and 8, 1972, that it believed the vaccine was extremely effective in controlling VEE in Texas in 1971. It set the following guidelines: all adult equines in the United States including pregnant mares at least 60 days pregnant which have not been previously immunized should be vaccinated with live attenuated VEE vaccine TC 83; foals of immune dams in high risk areas should be vaccinated when three months of age and again when weaned at approximately six months of age; previously unvaccinated pregnant mares in high risk areas should be vaccinated regardless of the stage of the pregnancy; foals not in high risk areas should be vaccinated initially at six months of age or with the waning of the maternal immunity when that time is known; the highest priority is recommended for the prompt initial vaccination of all horses in the Gulf Coast states and the states adjoining Texas and Mexico; the revaccination of adult horses in states other than those in which the disease is known to have occurred is not deemed necessary within 18 months of the original vaccination.

The problem of the lack of sentinel animals is a real one but the solution is now at hand. It is the availability of the wild burro. There are in southern California 3,380 wild unvaccinated burros, and in Arizona 7,510. In California, most of the burros are located in Inyo, San Bernardino and Imperial Counties. The China Lake Naval Base has asked Federal and State agencies for permission to remove 200 animals to avoid starvation because the burros have outproduced their forage and are in danger of destroying the desert plant life unless their numbers are reduced. If a properly handled humane approach to the problem can be proposed, I believe that those interested in preserving the burros would not object. I propose a pilot program which would initially involve the relocation of 10 or 20 burros to the Tule Elk Preserve in Kern County. A separate section could be provided, or the elk and burros could run on the same range. If this proposed pilot program can be successfully worked out with the State Department of Fish and Game, the State Park Department, the Federal Departments of Agriculture and Interior, a new source of federal animals would be available for responsible agencies that will be invaluable to these agencies in disease control. This will solve the sentinel animal problem and also provide relief for the overstocked desert ranges now being depleted by the burros. I earnestly request the Executive Board of this organization to study this proposal and offer its suggestions for implementing this plan.

CONCLUSIONS.—In a studied reflection based on personal observations and involvement in many levels of the 1971 VEE epizootic in Texas, Mexico and California, there was only one saving grace — the TC 83 vaccine. The spraying for mosquitoes and the public awareness from press reports and other media probably helped keep the number of human cases down to 88. The quarantine helped some. VEE was not as glamorous, nor was it vaccinated for with enough vigor, or we would not have had the 450 cases reported in 1972. The same might be said about Eastern Equine Encephalitis, in which 30 horse cases were reported in 1972, indicating that the vaccination program may not have been sufficiently wide spread. I believe we have the means to con-

trol all 3 diseases in the horse — VEE, Western equine encephalitic (WEE), and Eastern equine encephalitis (EEE), but do we have the desire to control them? We must develop and support in this organization similar groups of people dedicated to disease control, a fresh new outlook that includes a task force of active participants at world-wide conferences on infectious diseases. This should also include study teams to work with exotic diseases in their native habitats. We cannot wait until another arbovirus threatens our livestock or human populations before we act. African horse sickness could well be the next problem we face and we cannot afford the luxury of playing “catch-up” in disease control.

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# SURVEILLANCE FOR ARTHROPOD-BORNE VIRUSES AND DISEASE BY THE CALIFORNIA STATE DEPARTMENT OF PUBLIC HEALTH

Richard W. Emmons<sup>1</sup>, Gail Grodhaus<sup>2</sup> and Dario Cappucci<sup>3</sup>

Extensive surveillance for human and equine arbovirus encephalitis and for the causative viruses was carried out during 1972, as in previous years, but consisted of an even larger effort than in 1971 because of the continued threat that Venezuelan equine encephalitis (VEE) might spread northward through Mexico and reach California. As usual, there was collaboration by many persons in this Department and in other local, county, state and federal agencies, including the Schools of Public Health of the University of California at Berkeley and Los Angeles, the University of California at Davis, the State Department of Food and Agriculture, the United States Department of Agriculture, county health departments, mosquito abatement districts, and the Center for Disease Control. The results of the surveillance program can only be briefly summarized here.

At least 797 persons suspected of having an arbovirus infection were screened serologically by the State Viral and Rickettsial Disease Laboratory or the four county health department laboratories which have begun arbovirus testing (Table 1). We anticipate that local laboratories will increasingly assist in this surveillance. There were two human cases of VEE (exposures in Mexico), three cases of western equine encephalitis (WEE), and five cases of St. Louis encephalitis (SLE) detected (Table 2). All cases recovered, and no serious sequelae were reported. In addition, three non-fatal cases of dengue were detected in persons exposed overseas. Details of these cases are shown in Table 3. Cases were confirmed serologically by complement-fixation, hemagglutination-inhibition, neutralization, and indirect fluorescent antibody (IFA) tests. The value and specificity of the IFA test and of a metabolic inhibition test (neutralizing antibody) in cell cultures, developed in the Viral and Rickettsial Disease Laboratory, has been further confirmed.

There were 68 clinically suspect cases of encephalitis in equines reported to the Department in 1972 (Table 4), but vaccine-induced or naturally-acquired antibodies already present in sera prevented a definitive diagnosis in nearly all cases. Tissue samples from 31 equines were tested in suckling mice, with negative results. In addition, tissue samples from 13 sentinel hamsters, and over 100 wildlife specimens were tested with negative results. Serologic sampling of over 300 rodents or other mammals trapped in San Diego, Los Angeles, Imperial, Riverside, Stanislaus, and Colusa counties, by metabolic inhibition test for WEE and VEE antibodies, revealed only low titers (1:4 to 1:8) in a few animals, the significance of which is uncertain.

A total of 6,336 mosquito pools were tested in suckling mice or duck embryo primary cell cultures (Tables 5 and 6). There were 180 viruses isolated, as shown in detail (Table 7) and by county and month in Table 8. A number of the "unidentified viruses" appear to be Bunyamwera group agents, but they have not yet been fully characterized. No isolates of VEE virus, either vaccine or wild strains, were made. The majority of mosquito pools were tested in suckling mice, but duck embryo primary cell cultures were also used this year for the first time, to supplement the testing effort. Direct fluorescent antibody staining was of great value in identifying WEE, SLE, and Turlock virus isolates. The method is rapid, specific and accurate, resulting in considerable saving of time and materials as compared with neutralization tests to identify isolates.

The surveillance program during 1972 also involved several statewide news alerts to inform the public and the medical community; special technical bulletins sent to veterinarians, physicians and laboratories; a monthly progress report (June through November) to a large number of interested individuals; and a weekly report of mosquitoes tested and viruses isolated, which was sent to vector control specialists in the field. In summary, we feel that a good job of surveillance was done within the limits of budget, personnel and time restrictions. Unfortunately, we cannot rest on our laurels. Venezuelan equine encephalitis remains a threat; western and St. Louis encephalitis viruses are still widely endemic in California; other arboviruses which we know occur in the state may be, or become, capable of causing human illness; mosquitoes increasingly are developing resistance to insecticides; a large percentage of the increasing population of California presumably is susceptible to the viruses; and a banner year for excess water and mosquito breeding sites is clearly underway. Although we are faced with limited budgets and the probability of reduced research support, it will be as important as ever to maintain an efficient surveillance system, expand local laboratory capabilities, develop ecologically sound mosquito control methods, and continue trying to understand the basic virus cycles and how to predict and prevent arbovirus epidemics.

## Acknowledgement

The assistance and cooperation of many staff members of the Viral and Rickettsial Disease Laboratory, the Bureau of Vector Control and Solid Waste Management, other units of the California State Department of Public Health, all too numerous to mention individually, as well as of local mosquito abatement districts and other local, county, state and federal agencies, in carrying out the surveillance program are gratefully acknowledged.

<sup>1</sup>Viral and Rickettsial Disease Laboratory and Bureau of Communicable Disease Control.

<sup>2</sup>Bureau of Vector Control and Solid Waste Management.

<sup>3</sup>Veterinary Section.

Table 1.—Humans tested serologically for mosquito-borne arboviruses by the Viral and Rickettsial Disease Laboratory, California State Department of Public Health and by county health department laboratories, by county of residence and month of illness onset — California, 1972.

COUNTY	TOTAL	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	UNK
California	797	4	2	5	12	63	89	110	133	106	82	88	15	88
Alameda	26	1	—	—	—	2	3	—	2	1	3	6	—	8
Butte	14	—	—	—	—	—	3	2	1	2	2	3	—	1
Colusa	1	—	—	—	—	—	—	1	—	—	—	—	—	—
Contra Costa	23	—	—	—	1	2	5	—	3	2	4	3	—	3
Fresno**	53	—	1	—	1	—	4	15	7	10	4	5	4	2
Glenn	6	—	—	—	—	—	—	1	3	2	—	—	—	—
Humboldt	1	—	—	—	—	—	—	1	—	—	—	—	—	—
Imperial	3	—	—	—	—	—	—	—	1	1	—	—	—	1
Kern	36	1	—	—	—	3	4	2	2	2	5	3	—	14
Kings	6	—	—	—	—	—	1	—	—	—	1	1	—	3
Lake	3	—	—	—	—	—	—	—	—	3	—	—	—	—
Los Angeles*	70	—	—	—	3	6	13	9	10	6	5	7	7	4
Marin	12	—	—	1	1	—	1	2	3	—	—	2	—	2
Mendocino	1	—	—	—	—	—	—	—	1	—	—	—	—	—
Merced	2	—	—	—	—	—	—	—	—	—	—	—	—	2
Modoc	1	—	—	—	—	—	—	—	—	1	—	—	—	—
Monterey	15	—	—	—	—	—	1	5	2	2	—	2	—	3
Napa	4	—	—	—	—	1	—	1	1	—	—	—	—	1
Nevada	2	—	—	—	—	—	—	—	1	—	1	—	—	—
Orange	1	—	—	—	—	—	—	—	—	—	—	1	—	—
Placer	3	—	—	—	—	—	1	—	1	—	—	1	—	—
Riverside	7	—	—	—	—	—	3	2	2	—	—	—	—	—
Sacramento****	45	—	—	—	1	2	4	2	9	10	5	5	3	4
San Bernardino	19	—	—	—	—	1	—	5	3	3	2	3	—	2
San Diego***	61	—	—	—	1	7	6	11	11	12	9	3	—	1
San Francisco	79	—	—	1	1	5	8	15	13	12	10	4	—	10
San Joaquin	25	—	—	—	—	2	4	3	5	1	3	2	—	5
San Luis Obispo	6	—	—	—	—	—	1	1	2	—	—	—	—	2
San Mateo	38	1	—	—	—	3	3	6	7	2	5	10	—	1
Santa Barbara	21	—	1	1	—	4	1	2	8	1	1	—	—	2
Santa Clara	100	—	—	2	1	15	12	12	20	13	12	9	—	4
Santa Cruz	14	1	—	—	—	1	2	—	2	—	1	3	—	4
Shasta	3	—	—	—	—	—	—	—	—	3	—	—	—	—
Sierra	1	—	—	—	—	—	—	—	—	1	—	—	—	—
Siskiyou	1	—	—	—	—	—	—	—	—	—	1	—	—	—
Solano	14	—	—	—	—	3	1	2	—	6	1	1	—	—
Sonoma	13	—	—	—	—	1	—	—	1	1	1	7	—	2
Stanislaus	11	—	—	—	1	—	—	1	3	3	2	—	—	1
Sutter	3	—	—	—	—	1	1	1	—	—	—	—	—	—
Tehama	9	—	—	—	—	1	3	1	—	1	2	1	—	—
Tulare	14	—	—	—	1	1	1	3	1	2	1	2	—	2
Ventura	10	—	—	—	—	—	2	—	3	—	—	3	1	1
Yolo	18	—	—	—	—	2	1	4	5	3	1	1	—	1
Out of State	1	—	—	—	—	—	—	—	—	—	—	—	—	1
Unknown	1	—	—	—	—	—	—	—	—	—	—	—	—	1

\* Tested by County Health Department Laboratory (includes 9 patients tested by State VRDL)

\*\* Tested by County Health Department Laboratory (includes 18 patients tested by State VRDL)

\*\*\* Tested by County Health Department Laboratory (includes 3 patients tested by State VRDL)

\*\*\*\* Tested by County Health Department Laboratory (includes 19 patients tested by State VRDL)



Table 2.—Human cases of arthropod-borne encephalitis — California, 1950-1972.

YEAR	TOTAL	WEE	SLE	DEATHS
1950	157	88	69	2 — (1 WEE, 1 SLE)
1951	55	22	33	—
1952	420	375	45	10 — (9 WEE, 1 SLE)
1953	36	14	22	1 — (SLE)
1954	121	22	99	2 — (SLE)
1955	9	6	3	—
1956	21	14	7	—
1957	26	3	23	1 — (SLE)
1958	53	37	16	—
1959	42	2	40	1 — (SLE)
1960	13	1	12	—
1961	10	2	8	1 — (WEE)
1962	21	5	16	1 — (SLE)
1963	15	3	12	—
1964	12	10	2	—
1965	10	9	1	—
1966	17	9	8	—
1967	15	7	8	—
1968	15	11	4	—
1969	5	—	5	—
1970	2	—	2	—
1971	5*	3	2	—
1972	8**	3	5	—

\*In addition, 1 case of VEE confirmed, in traveler to Mexico.

\*\*In addition, 2 cases of VEE confirmed, in travelers to Mexico.

Table 3.—Human cases of Western, St. Louis, Venezuelan and Dengue Virus Infections in California, 1972.

Ident. & presumed exposure	Clinical Course	Test	Antibody Titers		Remarks	
			Acute	Convalescent		
1. T.P., 25, male, Chula Vista, San Diego Co. During month before onset, visited Borrego Springs (San Diego Co.) & Yuma Arizona, but no known mosquito exposure. During 2 weeks before onset, visited Lima, Cuzco, and Machu Picchu, Peru; & Rio de Janeiro, Brazil, with heavy mosquito exposure. (Arrived Brazil 3 days before onset).	Onset Jul/14/72 Severe HA, nausea, vomiting, fever and sweats; spinal tap. Had Yellow Fever vaccination Jun/21/72.	SLE-CF SLE-HAI SLE-IFA SLE-NI  YF-IFA	Jul/24/72 <1:4 <1:30 1:16 qns  1:128	Aug/9/72 1:8 >1:160 1:128 qns  >1:256	CF tests for VEE, WEE, Dengue 1 & 2 <1:8; HAI tests for WEE, VEE, Pow, Modoc, CEV, Turlock, Main Drain, BW, Lokern <1:10	
2. J.S.C., 22, male, Santa Cruz, Santa Cruz Co. Exposure in Santa Cruz, San Blas, & Puerto Vallarta in Nayarit & Jalisco State, Mexico; heavy mosquito exposure.	Onset Aug/9/72 HA, fever, chills, severe muscle aches	VEE-CF VEE-HAI VEE-IFA VEE-MI WEE-CF WEE-HAI WEE-IFA	Aug/11/72 1:8 1:20 1:128 1:32 — <1:10 <1:8	Aug/13/72 — 1:20 — — <1:10 —	Aug/31/72 1:8 1:20 1:128 1:64 <1:8 <1:10 <1:8	No virus isolated in s.m. from acute serum. No hx of VEE vaccination; HAI tests <1:10 for SLE, POW, Modoc, CEV, Turlock, Main Drain, BW & Lokern viruses. Presumptive positive VEE.
3. S.S., 4 month (Apr/17/72), female. Madera, Madera County	Onset Aug/22/72 Fever, convulsions, LP 30 cells	WEE-CF WEE-HAI WEE-IFA WEE-MI VEE-HAI VEE-CF VEE-IFA VEE-MI	Aug/29/72 <1:8 1:1280 1:64 1:16 <1:10 <1:8 <1:4 qns	Sept/21/72 1:64 1:2560 1:256 1:128 <1:10 <1:8 <1:4 <1:4	Hospitalized in Fresno WEE + mosquito pools near residence. CF tests <1:8 for measles, mumps, herpes, SLE	
4. B.K., ?, male. Entomologist, Los Angeles County. Exposure in So. Sinaloa State, Mexico.	Onset Aug/24/72	No Details	No Details	No Details	VEE confirmed by virus isolation from acute serum and by serology	
5. S.A., 14, male, Fresno. Fresno County. Camped by San Joaquin River 2 weeks prior to onset. Mosquito bite near eye.	Onset Sept/8/72 HA, fever, confusion, drowsy, then stupor, stiff neck.	WEE-CF WEE-HAI WEE-IFA  WEE-MI VEE-CF VEE-HAI VEE-IFA VEE-MI	8/11/72 <1:8 1:10, 240 1:128 repeat 1:64 1:16 <1:8 <1:10 qns qns	8/27/72 1:128 1:5, 120 1:512 1:256 qns <1:8 <1:10 qns qns	12/8/72 1:16 — — 1:64 1:64 <1:8 — <1:4 <1:4	
6. H. H., 35, male, Fresno, Fresno County. Mosquitoes abundant around home.	Onset 8/10/72 HA, malaise, fever 101-103 F, stiff neck, drowsy.	WEE-CF WEE-HAI WEE-IFA WEE-MI  VEE-CF VEE-HAI VEE-IFA VEE-MI	8/21/72 1:8 1:2560 1:128 1:8 <1:8 <1:10 <1:4 <1:4	10/3/72 1:32 1:10, 240 1:512 1:16 — <1:8 <1:10 <1:4 <1:4	12/8/72 1:32 1:5, 120 1:256 — — <1:10 <1:4 —	CF tests <1:8 for adenovirus, M. pneumonia, Q fever, mumps, SLE; Herpes CF titer stationary at 1:32
7. J. M., 11, male, Los Molinos, Tehama Co.	Onset Sept/13/72 stiff neck, high fever, 54 cells in CSF	SLE-CF SLE-HAI SLE-IFA SLE-NI	8/1 (?) <1:4 1:640 1:32 1.7	8/27/72 1:16 1:640 1:128 3.0	CF test <1:8 for measles, herpes, VEE, and WEE; mumps CF 1:8-1:8	

Table 3.—Continued.

Ident. & presumed exposure	Clinical Course	Test	Antibody Titers		Remarks
			Acute	Convalescent	
8. H. H., 25, male, Davis, Yolo Co.	Onset Sept/14/72 HA, fever, obtundation alternating with marked agitation	SLE-CF SLE-HAI SLE-IFA SLE-NI	Sept/18/72 <1:4 1:320 1:32 0.5	Sept/27/72 1:32 1:2,560 1:256 3.7	CF titers neg. or stationery for adeno- virus, mycoplasma, Q fever, mumps, herpes, VEE, WEE, measles, No virus isolated from TW or CSF.
9. R.L.S., 55, Male, Earlimart, Tulare County.	Onset Sept/22/72; tremors, chills, sweats, stiff neck, unstable gait, dis- oriented	SLE-CF SLE-HAI SLE-IFA SLE-NI	Sept/29/72 1:16 1:2560 1:512 1.9	10/10/72 1:64 1:2560 1:1024 3.8	Hospitalized Fresno; CF tests (1:8 for VEE, WEE; stationary titers for mumps (1:8), herpes (1:64) & measles (1:16)
10. H.M., 17, Male, Delano, Kern County.	Onset 10/5/72, admi. to hosp. 10/7; sudden, severe HA, fever to 106, seizure stiff neck, 69 WBC in CSF	SLE-CF SLE-HAI SLE-IFA SLE-IFA SLE-NI	10/9/72 1:16 - 1:32 1:64 qns	10/29/72 - - 1:128 1:256 1.0 12/5/72 1:16 - - 1:256 3.0	
11. S.M., 20, Female, Santa Cruz, Santa Cruz County	Jan/13/72; fever, chills jnt. pains, rash, eye pain	Dengue 1-CF Dengue 2-CF	2/7/72 1:128 1:256	3/21/72 1:32 1:64	Exposure in western Samoa, Upolu Island Dec/31/72 to Jan/22/72
12. A.G., 50, female, San Rafael, Marin County  HAI tests -- Courtesy CDC Lab  Dengue 1     <1:10   1:10     1:10 Dengue 2     <1:10   1:20     1:20 Dengue 3     <1:10   1:10     1:10 Dengue 4     <1:10   1:20     1:20 Yellow Fever <1:10   1:20     1:20 SLE           <1:10   <1:10   <1:10	1/19/72; fever, myalgia rash	Dengue 1-CF Dengue 2-CF Dengue 2-IFA Dengue 1-PRNT Dengue 2-PRNT Dengue 3-PRNT Dengue 4-PRNT	1/25/72 <1:4 <1:4 <1:8 <1:10 1:10 <1:10 <1:10	2/4/72     3/8/72 <1:4     1:4 1:4     1:8 1:64  <1:10 1:75 <1:10 <1:10	Exposure in Haiti 7 days p.t.o.  PRNT courtesy of Dr. Phil Russell  HAI titers <1:10 for EEE, WEE, CEV; CF titers (1:8 for Dengue 3 & 4, Yellow fever, SLE.
13. J.C., 29, Female, Los Angeles. Never had YF vaccine.	Onset 7/27/72 while in Hong Kong; severe HA, myalgia, malaise, fever 103; pruritic rash Aug/2	Dengue 1-CF Dengue 2-CF SLE-IFA Yellow Fever IFA	Aug/2/72 1:128 1:256 1:4096 1:8192	Aug/17/72 1:128 1:256 1:4096 1:4096	Tour mid-July of Hawaii, Japan, Thailand, Singapore Hong Kong; heavy mos- quito exposure.

Several other clinically suspect cases of dengue in recent travelers to the Pacific area were reported, but the diagnosis could not be confirmed because convalescent sera were not submitted.

Table 4.—Suspected clinical cases of arbovirus encephalitis in equines, by county and month, for California, 1972.

COUNTY	MONTH OF ONSET													Totals
	Jan.	Feb.	March	April	May	June	July	August	Sept.	Oct.	Nov.	Dec.	Unknown or undetermined	
Totals	2	3	1	2	4	4	14	9	10	4	2	1	12	68
Alameda									1				1	1
Contra Costa													3	1
Fresno	1				1		1		2					8
Glenn								1	1				1	2
Imperial						1		1	1					4
Inyo								2						2
Kern*	1			1			3						1	5
Los Angeles														1
Madera				1			1							2
Mendocino					1	1	1							3
Merced							2		1	1				5
Nevada		1						1					3	5
Orange								1						1
Placer								1						1
Riverside											1		1	2
Sacramento					1							1	1	2
San Diego							1		1	1				3
San Francisco		1												1
San Joaquin		1	1				2			1				5
San Luis Obispo									1					1
Santa Barbara					1	1								2
Santa Clara							3							3
Stanislaus						1			1					2
Sutter										1				1
Tulare								2	1	1				3
Tuolumne											1			1
Yolo													1	1

\*Only one case could be confirmed serologically as WEE: a 3-year old gelding from Kern County, with onset July 24, 1972, and no history of vaccination. Interpretation of serologic results on other cases was difficult due to previous immunizations with WEE, VEE, or EEE vaccines.

Table 5.—Numbers of mosquitoes tested by County and Month, California, 1972, by the Viral and Rickettsial Disease Laboratory.\*

County	January	February	March	April	May	June	July	August	September	October	November	December	Total
Colusa					178 (10)	2,250 (47)	2,896 (80)	10,561 (261)	817 (28)	616 (16)			17,318 (442)
Fresno				5 (3)	97 (10)	501 (20)	621 (25)	1,077 (27)	4,766 (114)	9 (3)			7,076 (202)
Imperial	78 (7)	32 (8)	198 (18)	691 (37)	4,569 (124)	6,274 (187)	2,523 (94)	2,277 (82)	3,182 (127)	16,283 (504)	5,939 (216)	2,252 (100)	44,298 (1,504)
Inyo						55 (18)	268 (8)						323 (26)
Kern					517 (14)	932 (27)	1,961 (48)	854 (23)	1,170 (32)	82 (7)			5,516 (151)
Kings				26 (3)	93 (9)	184 (9)	117 (6)	59 (3)	210 (8)				689 (38)
Lassen						1,466 (36)		900 (20)	91 (4)				2,457 (60)
Los Angeles						116 (7)		236 (20)	143 (9)				495 (36)
Madera				24 (5)	264 (36)	752 (35)	1,849 (47)	1,801 (41)	2,048 (55)	982 (49)			7,720 (268)
Merced					1,309 (50)	1,049 (39)	1,739 (53)	7,210 (160)	2,383 (60)				13,690 (362)
Modoc						2,635 (61)	132 (5)	2,527 (57)					5,294 (123)
Orange								1,374 (43)					1,374 (43)
Placer						294 (13)	704 (32)	1,964 (51)	1,850 (64)	493 (12)			5,305 (172)
Plumas							1,342 (30)	2,349 (51)					3,691 (81)
Riverside						655 (29)	357 (13)	6,547 (227)	2,278 (76)				9,837 (345)
Sacramento						139 (7)	278 (12)		337 (12)	546 (21)			1,300 (52)
San Bernardino						10 (2)	229 (13)	268 (10)	250 (15)				757 (40)

Table 5.--(Continued.)

County	January	February	March	April	May	June	July	August	September	October	November	December	Total
San Diego	13 (2)	133 (7)	63 (9)	812 (21)	2,333 (72)	5,555 (150)	4,014 (102)	4,598 (162)	1,987 (70)	1,519 (61)	609 (37)	292 (10)	21,928 (703)
San Luis Obispo							48 (13)	112 (10)	141 (13)				301 (36)
Santa Barbara						24 (4)	57 (5)	139 (9)		133 (10)			353 (28)
Shasta			245 (35)	212 (35)	259 (30)	1,536 (106)	3,334 (89)	1,649 (51)	271 (17)				17,506 (363)
Siskiyou							2,006 (63)	818 (31)	233 (8)				3,057 (102)
Stanislaus						440 (22)	1,273 (40)	993 (36)	704 (28)				3,410 (126)
Sutter						325 (8)	1,152 (34)	659 (20)	1,112 (33)	1,256 (29)			4,504 (124)
Tehama			254 (36)	91 (42)	186 (30)	1,772 (70)	2,190 (66)	2,162 (65)	1,553 (57)				8,208 (366)
Tulare				57 (5)	8 (4)	14 (6)	16 (5)	3,404 (79)	2,427 (59)	2,573 (59)			8,499 (217)
Ventura						15 (4)	34 (7)	28 (5)	27 (12)				104 (28)
Yolo						1,537 (36)		2,298 (51)	318 (9)	1,048 (31)			5,201 (127)
Yuba							106 (3)			600 (12)			706 (15)
Yuma, Arizona	66 (3)	5 (3)	144 (8)		732 (23)	721 (18)	320 (13)	230 (10)	668 (27)	710 (38)	56 (3)	168 (10)	3,820 (156)
<b>Totals</b>	<b>157 (12)</b>	<b>170 (18)</b>	<b>904 (106)</b>	<b>1,918 (151)</b>	<b>10,545 (412)</b>	<b>29,196 (943)</b>	<b>29,353 (916)</b>	<b>57,362 (1,613)</b>	<b>28,966 (937)</b>	<b>26,850 (852)</b>	<b>6,604 (256)</b>	<b>2,712 (120)</b>	<b>194,737 (6,336)</b>

\*Total mosquitoes (pools) tested.

Table 6.—Numbers of mosquitoes tested, by species and month, California, 1972 by the Viral and Rickettsial Disease Laboratory.\*

Species	January	February	March	April	May	June	July	August	September	October	November	December	Total
<i>Culex tarsalis</i>	85 (4)	10 (4)	418 (41)	527 (36)	3,549 (124)	15,226 (403)	16,697 (454)	25,783 (665)	>14,156 (385)	10,604 (289)	694 (43)	1,040 (45)	>88,789 (2,493)
<i>erythrothorax</i>	62 (3)	138 (6)	195 (9)	777 (18)	3,530 (83)	4,964 (122)	>2,182 (55)	>2,828 (79)	1,803 (53)	1,628 (58)	2,178 (66)	1,367 (40)	>21,652 (592)
<i>peus</i>			1 (1)	53 (4)	60 (10)	1,100 (68)	908 (44)	1,013 (65)	1,008 (57)	29 (7)	26 (2)		4,198 (258)
<i>pipiens</i>	5 (2)		4 (3)	104 (9)	1,051 (34)	1,920 (63)	746 (34)	2,295 (79)	>2,766 (104)	3,069 (102)	1,091 (38)	81 (12)	>13,132 (480)
<i>apicalis</i>			1 (1)										1 (1)
<i>thriambus</i>						3 (1)							3 (1)
<i>Aedes melanimon</i>					942 (38)	>1,646 (62)	1,970 (66)	5,876 (150)	1,593 (51)	640 (20)			>12,667 (387)
<i>vexans</i>			8 (3)	127 (7)	924 (29)	2,262 (67)	2,266 (64)	2,653 (104)	2,772 (80)	3,882 (116)	2,253 (54)	13 (4)	17,160 (528)
<i>dorsalis</i>		16 (3)	18 (5)	9 (5)	57 (9)	553 (22)	920 (34)	519 (18)	185 (11)	38 (19)	23 (5)		2,338 (131)
<i>nigromaculis</i>				59 (3)	53 (10)	409 (17)	1,761 (53)	4,847 (110)	719 (31)	268 (14)			8,116 (238)
<i>sierrensis</i>			57 (6)	104 (17)	117 (9)	75 (9)	8 (2)	1 (1)					362 (44)
<i>increpitus</i>					89 (4)	3 (1)							92 (5)
<i>squamiger</i>													
<i>taeniorhynchus</i>							175 (8)	500 (17)	28 (3)	4 (1)	2 (1)		709 (30)
<i>Anopheles freeborni</i>			171 (16)	35 (17)	21 (14)	>222 (25)	1,016 (42)	2,881 (76)	1,684 (62)	3,246 (71)			>9,276 (323)
<i>occidentalis</i>					1 (1)	42 (5)	37 (7)	1 (1)					81 (14)
<i>franciscanus</i>			1 (1)	3 (2)	6 (4)	40 (16)	234 (16)	216 (23)	133 (18)	126 (9)	4 (3)		763 (92)
<i>punctipennis</i>			3 (3)	9 (4)	46 (11)	>259 (15)	2 (1)	289 (14)	5 (4)	5 (2)			>618 (54)
<i>Culiseta incidens</i>		1 (1)	>15 (7)	91 (19)	41 (12)	>114 (18)	38 (7)	44 (6)	61 (4)	17 (5)	9 (3)		>431 (82)
<i>inornata</i>	5 (3)	5 (4)	11 (9)	20 (10)	58 (20)	249 (21)	270 (19)	280 (13)	110 (12)	34 (10)	324 (41)	211 (19)	1,577 (181)
<i>particeps</i>			1 (1)			3 (1)	6 (1)	18 (2)	12 (2)	1 (1)			41 (8)
<i>Psorophora confinis</i>						106 (7)	117 (9)	7,309 (185)	1,931 (60)	3,223 (122)			12,686 (383)
<i>signipennis</i>								9 (5)		36 (6)			45 (11)
<b>Totals</b>	<b>157 (12)</b>	<b>170 (18)</b>	<b>&gt;904 (106)</b>	<b>1,918 (151)</b>	<b>10,545 (412)</b>	<b>&gt;29,196 (943)</b>	<b>&gt;29,353 (916)</b>	<b>&gt;57,362 (1,613)</b>	<b>&gt;28,966 (937)</b>	<b>26,850 (852)</b>	<b>6,604 (256)</b>	<b>2,712 (120)</b>	<b>&gt;194,737 (6,336)</b>

\*Total mosquitoes (pools) tested.

Table 7.—Viral isolates from mosquito pools, by the Viral and Rickettsial Disease Laboratory, California State Department of Public Health, 1972.

Identifying number	County	Place	Date Collected	Species	Number In Pool	Isolate
V1-306	Tehama	Woodson Bridge	June 13	<i>Culex tarsalis</i>	50	Turlock
V5-235	Imperial	Calexico	June 13	<i>C. tarsalis</i>	50	WEE
V5-242	Imperial	Calexico	June 13	<i>C. tarsalis</i>	50	SLE
V5-318	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	WEE
V5-323	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	WEE
LAV5-25	Yuma, Arizona	Morelos Dam	June 15	<i>C. tarsalis</i>	50	WEE
V5-238	Imperial	Calexico	June 13	<i>C. tarsalis</i>	50	SLE
V5-240	Imperial	Calexico	June 13	<i>C. tarsalis</i>	50	Turlock
V5-261	Imperial	Calexico	June 13	<i>C. tarsalis</i>	40	Turlock
V5-268	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	Turlock
V5-303	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
V5-311	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	Turlock
V5-320	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
V5-322	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
V5-348	Imperial	Calexico	June 14	<i>Aedes vexans</i>	50	Turlock
V5-354	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
V5-355	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
V5-356	Imperial	Calexico	June 14	<i>C. tarsalis</i>	50	SLE
LAV5-26	Yuma, Arizona	Morelos Dam	June 15	<i>C. tarsalis</i>	50	Turlock
LAV5-28	Yuma, Arizona	Morelos Dam	June 15	<i>C. tarsalis</i>	46	Turlock
LAV5-54	Imperial	Yuma Mission	June 16	<i>C. tarsalis</i>	49	Turlock
V1-494	Tehama	Corning	June 23	<i>C. tarsalis</i>	28	Turlock
V5-404	Riverside	Blythe	June 28	<i>C. tarsalis</i>	50	WEE
LAV5-110	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	WEE
V1-383	Modoc	Alturas	June 19	<i>C. tarsalis</i>	50	Unidentified
LAV5-96	Imperial	Seeley	July 11	<i>C. tarsalis</i>	44	WEE
LAV5-104	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	SLE
LAV5-107	Imperial	Seeley	July 11	<i>C. tarsalis</i>	32	WEE/SLE
LAV5-109	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	WEE
LAV5-114	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	SLE
LAV5-121	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	WEE
LAV5-122	Imperial	Calexico	July 11	<i>C. tarsalis</i>	50	SLE
LAV5-138	Imperial	Calexico	July 12	<i>C. tarsalis</i>	11	SLE
LAV5-141	Imperial	Calexico	July 12	<i>C. tarsalis</i>	6	WEE
LAV5-144	Imperial	Calexico	July 12	<i>C. tarsalis</i>	26	WEE
LAV5-139	Imperial	Seeley	July 12	<i>C. tarsalis</i>	4	SLE
LAV5-149	Imperial	Seeley	July 12	<i>C. tarsalis</i>	50	WEE
LAV5-150	Imperial	Seeley	July 12	<i>C. tarsalis</i>	10	SLE
LAV5-155	Imperial	Calexico	July 12	<i>C. tarsalis</i>	19	SLE
LAV5-156	Imperial	Calexico	July 12	<i>Culex pipiens</i>	21	SLE
LAV5-164	Imperial	Winterhaven	July 13	<i>C. tarsalis</i>	33	SLE
V2-636	Yolo	Knights Landing	June 28	<i>C. tarsalis</i>	50	Turlock
V2-058	Sutter	Pennington	July 19	<i>C. tarsalis</i>	50	Turlock
V4-339	Madera	Cottonwood Creek	July 12	<i>C. tarsalis</i>	50	WEE
V4-345	Madera	Cottonwood Creek	July 12	<i>C. tarsalis</i>	50	WEE
V2-683	Stanislaus	Newman	July 11	<i>C. tarsalis</i>	50	Turlock
V2-687	Stanislaus	Newman	July 11	<i>C. tarsalis</i>	50	Turlock
V4-363	Kings	Riverdale	July 18	<i>C. tarsalis</i>	50	WEE
V2-605	Yolo	Knights Landing	June 27	<i>C. tarsalis</i>	50	Unidentified
V2-753	Colusa	Colusa	July 26	<i>C. tarsalis</i>	50	SLE
V2-733	Colusa	Princeton	July 25	<i>C. tarsalis</i>	44	Turlock
V4-421	Kern	Taft	July 31	<i>C. tarsalis</i>	50	Turlock
V4-440	Kern	Taft	July 31	<i>C. tarsalis</i>	50	Turlock
V4-443	Kern	Taft	July 31	<i>C. tarsalis</i>	50	Turlock
V5-452	San Diego	San Ysidro	July 11	<i>A. taeniorhynchus</i>	15	Unidentified
V5-480	San Diego	San Ysidro	July 13	<i>A. taeniorhynchus</i>	50	Unidentified
V2-773	Sacramento	Robla	July 31	<i>Culex peus</i>	9	SLE
V1-763	Shasta	Fall River M.	July 24	<i>Culex tarsalis</i>	50	SLE
V2-873	Placer	Sheridan	August 9	<i>C. tarsalis</i>	50	Turlock
V4-235	Merced	San Luis Ranch	June 26	<i>C. tarsalis</i>	50	Unidentified



Table 7.--(continued)

Identifying Number	County	Place	Date Collected	Species	Number In Pool	Isolate
V4-475	Madera	Madera	August 7	<i>Culex tarsalis</i>	50	WEE
V4-473	Madera	Madera	August 7	<i>C. tarsalis</i>	50	WEE
V4-477	Madera	Madera	August 7	<i>C. tarsalis</i>	50	WEE
V4-478	Madera	Madera	August 7	<i>C. tarsalis</i>	50	WEE
V1-847	Tehama	El Camino	August 8	<i>C. tarsalis</i>	50	SLE
V1-877	Shasta	Cottonwood	August 10	<i>C. tarsalis</i>	50	SLE
V1-883	Tehama	Red Bluff	August 10	<i>Culex peus</i>	35	SLE
V1-885	Tehama	Moon School	August 10	<i>C. tarsalis</i>	29	SLE
LAV5-328	Imperial	Seeley	August 9	<i>C. tarsalis</i>	50	SLE
LAV5-329	Imperial	Seeley	August 9	<i>C. tarsalis</i>	7	SLE
LAV5-344	Yuma, Arizona	Imperial Dam	August 11	<i>C. tarsalis</i>	50	SLE
V4-501	Kern	Taft	August 8	<i>C. tarsalis</i>	45	WEE
LAV5-309	Imperial	Seeley	August 8	<i>C. tarsalis</i>	34	WEE
V4-382	Fresno	Clovis	July 25	<i>C. tarsalis</i>	16	Turlock
V4-472	Madera	Madera	August 7	<i>C. tarsalis</i>	50	Turlock
V1-696	Tehama	Red Bluff	July 13	<i>C. peus</i>	47	Unidentified
V2-632	Yolo	Knights Landing	June 26	<i>C. tarsalis</i>	50	Unidentified
V1-873	Shasta	Cottonwood	August 10	<i>C. tarsalis</i>	50	Turlock
V2-903	Stanislaus	Newman	August 15	<i>Culex pipiens</i>	7	Turlock
V2-905	Stanislaus	Newman	August 16	<i>C. tarsalis</i>	50	Turlock
V2-908	Stanislaus	Newman	August 16	<i>C. tarsalis</i>	50	Turlock
V2-912	Stanislaus	Newman	August 16	<i>C. tarsalis</i>	37	Turlock
V4-487	Madera	Pinedale	August 7	<i>C. tarsalis</i>	50	Turlock
V4-541	Tulare	Goshen	August 17	<i>C. tarsalis</i>	50	SLE
V4-510	Madera	Mendota	August 15	<i>C. tarsalis</i>	21	Turlock
V4-514	Merced	Merced	August 16	<i>C. tarsalis</i>	50	Turlock
V4-521	Merced	Planada	August 16	<i>C. tarsalis</i>	16	Turlock
V4-530	Tulare	Goshen	August 17	<i>C. tarsalis</i>	50	Turlock
V4-536	Tulare	Goshen	August 17	<i>C. tarsalis</i>	50	Turlock
V5-861	San Diego	San Ysidro	August 10	<i>C. tarsalis</i>	50	Turlock
V5-905	San Diego	Rancho Santa Fe	August 11	<i>C. tarsalis</i>	50	Turlock
V1-941	Tehama	Woodson Bridge	August 21	<i>C. tarsalis</i>	50	SLE
V1-944	Tehama	Woodson Bridge	August 21	<i>C. tarsalis</i>	50	SLE
V1-947	Tehama	Woodson Bridge	August 21	<i>C. peus</i>	50	SLE
V1-973	Shasta	Palo Cedro	August 25	<i>C. tarsalis</i>	10	SLE
V1-593	Tehama	Red Bluff	July 6	<i>C. tarsalis</i>	37	Unidentified
V2-678	Stanislaus	Newman	July 11	<i>C. tarsalis</i>	50	Unidentified
V2-694	Stanislaus	Newman	July 11	<i>C. tarsalis</i>	50	Unidentified
V4-301	Merced	Planada	July 6	<i>C. tarsalis</i>	50	Unidentified
LAV5-583	Los Angeles	Montebello	August 18	<i>C. peus</i>	17	Turlock
V2-074	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	SLE
V2-075	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	SLE
V2-076	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	SLE
V2-077	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	SLE
V2-082	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	SLE
V2-091	Yolo	Woodland	August 28	<i>C. tarsalis</i>	50	SLE
V2-113	Colusa	Colusa	August 29	<i>C. tarsalis</i>	50	Turlock
V2-120	Colusa	Colusa	August 29	<i>C. tarsalis</i>	50	SLE
V2-126	Colusa	Colusa	August 29	<i>C. tarsalis</i>	50	SLE
V2-244	Colusa	Colusa	August 31	<i>C. tarsalis</i>	11	SLE
V4-592	Tulare	Woodville	August 21	<i>C. tarsalis</i>	50	WEE
V4-599	Tulare	Tipton	August 21	<i>C. tarsalis</i>	50	WEE
V4-667	Madera	Trigo	August 29	<i>C. tarsalis</i>	50	WEE
V4-668	Madera	Trigo	August 29	<i>C. tarsalis</i>	50	WEE
V4-670	Madera	Trigo	August 29	<i>C. tarsalis</i>	50	WEE
V4-674	Madera	Trigo	August 29	<i>C. tarsalis</i>	28	WEE
V2-994	Colusa	Colusa	August 23	<i>C. tarsalis</i>	50	Turlock
V5-1269	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	50	WEE
V4-593	Tulare	Woodville	August 21	<i>C. tarsalis</i>	49	SLE
V4-617	Kern	Bakersfield	August 24	<i>C. tarsalis</i>	18	WEE

Table 7.—(continued)

Identifying Number	County	Place	Date Collected	Species	Number In Pool	Isolate
V4-672	Madera	Trigo	August 29	<i>C. tarsalis</i>	50	WEE
V4-675	Madera	Trigo	August 29	<i>C. pipiens</i>	12	SLE
V5-1045	Imperial	Calexico	September 5	<i>C. tarsalis</i>	42	Turlock
V5-1238	Imperial	Seeley	September 6	<i>C. tarsalis</i>	50	SLE
V5-1258	Imperial	Winterhaven	September 7	<i>C. tarsalis</i>	46	SLE
V5-1270	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	50	WEE
V5-1273	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	13	SLE
V5-1281	Yuma, Arizona	Yuma	September 8	<i>C. tarsalis</i>	50	SLE
V5-1303	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	12	SLE
V5-1307	Yuma, Arizona	Yuma	September 8	<i>C. tarsalis</i>	50	SLE
V5-1311	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	50	Turlock
V5-1312	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	50	SLE
V5-1313	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	50	SLE
V5-1315	Imperial	Winterhaven	September 8	<i>C. tarsalis</i>	44	SLE
V5-1318	Imperial	Winterhaven	September 8	<i>C. erythrothorax</i>	50	SLE
V5-9029	Riverside	Blythe	August 29	<i>C. erythrothorax</i>	39	SLE
V5-947	San Diego	Imperial Bch	September 5	<i>C. erythrothorax</i>	50	SLE
V2-426	Colusa	Colusa	September 26	<i>A. melanimon</i>	50	WEE
V2-1015	Yolo	Woodland	October 3	<i>A. freeborni</i>	50	SLE
V2-135	Colusa	Colusa	August 29	<i>A. freeborni</i>	50	Unidentified
V2-156	Sutter	Meridian	August 30	<i>A. freeborni</i>	50	Unidentified
V2-142	Colusa	Colusa	August 29	<i>A. freeborni</i>	50	Unidentified
V2-224	Colusa	Cortena	August 30	<i>A. freeborni</i>	50	Unidentified
V2-235	Colusa	Colusa	August 31	<i>A. freeborni</i>	21	Unidentified
V2-321	Placer	Rocklin	September 11	<i>C. peus</i>	18	Turlock
V2-331	Placer	Lincoln	September 12	<i>C. peus</i>	50	Unidentified
V2-887	Stanislaus	Newman	August 14	<i>C. tarsalis</i>	46	Turlock
V2-981	Colusa	Cortena	August 22	<i>C. tarsalis</i>	50	Unidentified
V4-523	Merced	Planada	August 16	<i>A. melanimon</i>	17	Unidentified
V4-524	Tulare	Goshen	August 17	<i>C. tarsalis</i>	50	Unidentified
V4-1125	Madera	Madera	October 2	<i>C. tarsalis</i>	50	WEE
V1-905	Siskiyou	Yreka	August 15	<i>C. tarsalis</i>	50	Turlock
V1-928	Siskiyou	Indian Tom L.	August 15	<i>C. inornata</i>	17	Unidentified
V1-1140	Tehama	Gerber	September 11	<i>C. tarsalis</i>	50	SLE
V1-1142	Tehama	Gerber	September 11	<i>C. peus</i>	30	SLE
V1-1161	Tehama	Gerber	September 13	<i>C. tarsalis</i>	50	SLE
V5-800	San Diego	Imperial Bch	August 8	<i>A. taeniorhynchus</i>	19	Unidentified
V5-807	San Diego	Imperial Bch	August 9	<i>A. taeniorhynchus</i>	4	Unidentified
V1-1104	Tehama	Red Bluff	September 1	<i>C. tarsalis</i>	17	SLE
V4-1126	Madera	Madera	October 2	<i>C. tarsalis</i>	29	SLE
V4-1141	Tulare	Woodville	October 5	<i>C. tarsalis</i>	13	SLE
V1-1102	Plumas	Indian Valley	August 30	<i>C. tarsalis</i>	37	Turlock
V2-296	Yolo	Knights Landing	September 6	<i>C. tarsalis</i>	50	Turlock
V4-839	Madera	Firebaugh	October 12	<i>C. tarsalis</i>	50	WEE
V4-1137	Madera	Trigo	October 3	<i>C. tarsalis</i>	20	WEE
V4-1375	Imperial	Calexico	October 3	<i>C. tarsalis</i>	32	Turlock
V5-2038	Imperial	Winterhaven	October 5	<i>C. tarsalis</i>	50	Turlock
V5-2045	Imperial	Winterhaven	October 5	<i>C. tarsalis</i>	50	SLE
V5-2240	Imperial	Winterhaven	October 6	<i>C. tarsalis</i>	50	Turlock
V4-956	Fresno	Kerman	September 20	<i>C. tarsalis</i>	50	WEE
V4-964	Fresno	Kerman	September 20	<i>C. tarsalis</i>	50	WEE
V4-1007	Fresno	Mendota	September 20	<i>C. tarsalis</i>	50	WEE
V2-106	Colusa	Colusa	August 29	<i>C. tarsalis</i>	50	Unidentified
V2-196	Colusa	Delavan	August 30	<i>C. tarsalis</i>	50	Unidentified
V2-422	Sutter	West Butte	September 26	<i>C. tarsalis</i>	50	WEE
V4-815	Madera	El Peco	September 7	<i>C. tarsalis</i>	50	WEE
V4-823	Tulare	Woodville	September 11	<i>C. tarsalis</i>	39	WEE
V5-2386	Imperial	Calexico	November 9	<i>C. tarsalis</i>	12	Turlock
V1-1105	Tehama	Red Bluff	September 1	<i>C. peus</i>	13	Unidentified

Table 8.--Viruses isolated from mosquitoes by the Viral and Rickettsial Disease Laboratory, California State Department of Public Health, by County and Month of Collection, 1972.

County	June	July	August	September	October*
Cotusa		SLE (1) Turlock (1)	SLE (8) - Unid. (7) Turlock (2)	WEE (1)	
Fresno		Turlock (1)		WEE (3)	
Imperial	SLE (8) - WEE (3) Turlock (6)	SLE (10) WEE (8)	SLE (2) - WEE (1)	WEE (2) - SLE (8) Turlock (2)	SLE (1) Turlock (3)
Kern		Turlock (3)	WEE (2)		
Kings		WEE (1)			
Los Angeles			Turlock (1)		
Madera		WEE (2)	WEE (10) - SLE (1) Turlock (3)	WEE (1)	WEE (2) SLE (1)
Merced	Unid. (1)	Unid. (1)	Turlock (2) - Unid(1)		
Modoc	Unid. (1)				
Placer			Turlock (1)	Turlock(1)-Unid(1)	
Plumas			Turlock (1)		
Riverside	WEE (1)		SLE (1)		
Sacramento		SLE (1)			
San Diego		Unid. (2)	Turlock (2)-Unid(2)	SLE (1)	
Shasta		SLE (1)	SLE (2) - Turlock(1)		
Siskiyou			Turlock(1) - Unid(1)		
Stanislaus		Turlock(2) Unid(2)	Turlock (5)		
Sutter		Turlock (1)	Unid (1)	WEE (1)	
Tehama	Turlock (2)	Unid (2)	SLE (6)	SLE (4) - Unid. (1)	
Tulare			SLE (2) - WEE (2) Turlock (2)-Unid(?)	WEE (1)	SLE (1)
Yolo	Turlock(1)-Unid(2)		SLE (1)	Turlock (1)	SLE (1)
Yuma, Arizona	Turlock(2) - WEE(1)		SLE(1)	SLE (2)	
<b>TOTALS</b>	SLE (8) WEE (5) Turlock (11) Unident. (4)	SLE (13) WEE (11) Turlock (8) Unident. (7)	SLE (24) WEE (15) Turlock (21) Unident. (13)	SLE (15) WEE (9) Turlock (4) Unident. (2)	SLE (4) WEE (2) Turlock (3)

\*In addition, (1) Turlock in November from Imperial County. Results of additional tests on unidentified viruses pending (WEE, SLE and Turlock viruses ruled out).

# LEGAL ASPECTS OF MOSQUITO CONTROL

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**INTRODUCTION.** Those of us in California mosquito control have no choice but to face the reality that we are an integral part of a changing world dedicated to ecology and the environment. Individually we should also recognize that we are environmentalists in the true sense of the word and that our personal contribution should be purposeful environment modification based on our professional competence in the field of mosquito prevention, rather than mere mosquito control.

I am not going to tell you a story of "Goldilocks and the Three Bears". However, I am going to tell you about "Goldilocks (ourselves) and the Three R's"; the three R's being: Resistance, Regulations, and Responsibility.

If we are to retain the "Goldilocks" title in the field of public service we had better very rapidly create a new image for ourselves consistent with the present trend. We must re-evaluate our purpose and make necessary adjustments to reinforce or regain a much-needed respect for our programs. Fortunately, as we analyze our position it is rather obvious that we potentially deserve the "Goldilocks" rating. After all, there is very little we can recommend which is not consistent with the best interests of agriculture, land utilization and water conservation. . . provided of course, our performance is consistent to these objectives.

Needless to say, our present plight has been caused by resistance of mosquitoes to insecticides, as well as regulatory changes which continue to restrict our use of chemicals. However, our greatest omission in the past has been our failure to properly assign responsibility to the mosquito producer. This has been due partly to our lack of adequate communication with the extreme water user. Our greatest fault is that we have measured our programs in terms of gallons of insecticide and acres sprayed, rather than in mosquito sources eliminated or reduced. We are now in the third decade of the insecticide approach and it is little wonder that the mosquito producer is unaware of his legal obligations; in fact, we have given most mosquito producers good reason to conclude that extreme water usage is both normal and acceptable.

Today, because of increasing resistance in many areas of the State, we find ourselves unable to continue what has been a normal repetitive spray service, that of controlling mosquitoes on great numbers of acres of private land. Consequently, we have had to resort to legal abatement on an

urgency basis and regrettably we find ourselves poorly prepared to take this step.

Because legal action taken by a single mosquito abatement district can reflect on the near sixty total agencies, as well as to set precedents, the CMCA Source Reduction Committee initiated a group meeting in 1972. A request was approved by the CMCA Board of Directors to hold an Administrative Seminar on Legal Aspects of Mosquito Prevention. This was held in Berkeley on the last two days of October with the underlying purpose of producing an awareness of the problem and hopefully to condition our thinking prior to this Annual Conference, wherein we hope that wisdom will prevail in charting our collective future programs.

We are fortunate to have many of the same participants today for our Forty-First Annual Conference. It is my hope that this distinguished group can further advise us how to continue in the "Goldilocks" contest; perhaps some may suggest a new tint or dye, or possibly a brand new wig.

**SUMMARY.** It is apparent that we in mosquito control do not enjoy the rosy position which we occupied when many of us were just getting our feet wet in this field of public service. It is also evident that there is no easy single way to secure a solution to our unsettled problems of tomorrow. Neither our law, nor the abatement process offer in themselves a satisfactory means to our desired end.

However, what we do know is that we are agents of the Public, operating in the field of public health. To me, this means above all else that we must recognize that our efforts should also be dedicated toward preventive medicine, . . . in our case, mosquito prevention. Interestingly, the first definition of "prevent" in the dictionary is "to keep from occurring".

This, of course, brings us back to where our panel began and the inference that we must competently meet the challenge of this changing world dedicated to ecology and the environment. The likelihood of accomplishment will be realized when we combine the three "R's" of present concern into the single "R" of RESPECT. This "R" must be won through a rededication of our programs, to down-to-earth, person to person, cooperative modification of the environment. Fortunately, we will find that the respect will be mutual.

## WHAT'S NEW IN SOURCE REDUCTION

George R. Whitten

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Time after time over the last 20 years I have heard the same question posed by operators, managers, and trustees, Source Reduction Committee meetings, conferences, and seminars. The question in one form or another:

"How do we reach the grower and get his cooperation in reducing the mosquito problem which he is producing?"

This question has been cussed and discussed always in a rather frustrated manner because of the elusive nature of the topic and our inability to find a satisfactory, usable answer that we can apply to our programs with reasonable success.

The insurmountable hurdle has always been the grower's preoccupation with his existing problems based on his priority concepts and many personal interests. For us to reach him with our mosquito problems and take priority over his other interests requires an approach with considerable urgency. Now we have this urgency, which may very well, in 1973, grow to an emergency rather than just an urgency.

In 1969 we made a major change in program direction at the Delta MAD. Person to person contact — with two persons representing Delta MAD. We deemphasized spray. We faced the obvious fact of resistance and began to evaluate the effects it would have on our future. Without a cheap, effective insecticide, our spray program was a thing of the past. Time was right for a move away from insecticides which I think we all recognize was an opportunistic program which contained little opportunity for reducing our mosquito problem in any constructive manner.

The time was right for a move into a much more comprehensive source reduction program which would define the problem and place the responsibility on the producer, where it belonged.

We have taken the responsibility for mosquito control on our own shoulders over the last 25 years by the development of an insecticide program. Now we are giving it back to the grower. We feel that the source reduction program we have conducted over the last 20 years certainly proves to the grower that the District has and is making every effort to constructively assist him to solve the mosquito problem which he is creating.

The next step was to impress the producers with the necessity for immediate action to at least reduce the problem to a minimum level. This is the phase we are working on at this time and I think with reasonable success.

We have had a source reduction program at Delta for 20 years but with a cheap effective insecticide available, the lever to motivate owners and operators of large acreages of irrigated pasture to voluntarily spend the monies necessary to properly level the land and install adequate irrigation and drainage systems was a study in futility. I concentrated my efforts on growers who had smaller producing areas but were more progressive and willing to work with us to eliminate standing water on their property. After all, how many

times can you contact the same unresponsive grower and discuss the same problem with him without reaching an impasse. It is obvious to us that he will not move without stronger motivation which will impress him with the importance of immediate action.

We have used newspapers and other mass media methods in the past and will continue to use them in the future for educating the general public. But to reach the grower the only effective method we have found is an eyeball to eyeball meeting.

In the fall of 1968 and spring of 1969 District Superintendent Robert E. Turner and I made personal contacts with our major mosquito producers. We explained the difficult mosquito control situation which we were both facing. Resistance was here and within 2 to 3 years we anticipated that our available insecticides would be relatively ineffective. We presented him with the alternative of changing his present operations which were causing mosquito problems or facing an angry public who we believed would demand that he make these changes.

The first recommendation which we made was to change the crop. Irrigated pasture is our number one potential for mosquito production and the most effective method of reducing the source is to change the crop.

Many soils will not grow anything but pasture and these of course are among our worst problems. Many may not be irrigated without producing mosquitoes. Dr. Charles Schaefer of U. C. Fresno is conducting studies of these soil types at this time. We hope that, whether the results are positive or negative, they will be useful. We have made other suggestions, such as increased attention to water management, re-leveling the pastures and installing drainage, but where the soil problem is severe the monetary returns are poor, the incentive to invest in improvements is negative. As I mentioned, our change in direction occurred in 1969. We continued these contacts with these same growers through 1970 and 1971, reviewing the previous year's records and appealing to the grower to help to alleviate the problem which he was creating.

These contacts did impress some growers who instituted changes that improved the situation. In 1972 we began a program of citing to the Delta MAD Board of Trustees the growers that had not made a move. This was done very selectively and in a very friendly manner but the Board was also very firm in stating its position and the burden was on the producer. At this time we have the attention of the growers who have been cited plus many who had the word through the grapevine — grower to grower contact. We made more progress with the large pasture growers in 1972 than over the preceding 20 years. Whether we can hold their attention and make more improvements in 1973 we are uncertain but we have every intention of continuing the program in its present form. We don't feel that this program we

are using is strictly a legal approach — it is a forced education. The District has bent over backward or as one of our more vociferous auto dealers in the Valley says “I will stand on my head to make you a deal”. We are literally standing on our head to be sure that each grower has every opportunity to express himself to the staff when we contact him personally and to the Board when he is cited into a meeting and shown slides of his property.

I would like to show some of the slides we use at these hearings. I have chosen the first grower cited into the Board. He also has been at the top of our cost list for a number of years when we were spraying all sources routinely. In 1969 the basic airplane and insecticide costs were \$6,000. and if we multiply this by five which is the factor we use for all overhead, maintenance of equipment, etc., the cost comes to \$30,000.

We did \$1,300 worth of source reduction work for this grower after his citation this last summer; this of course was only a drop in the bucket when compared with the overall problem but it is a step in the right direction. He also paid \$130 in spray costs assessed against him.

The next example is a commercial plant producing stains and paints. This problem consisted of only seven acres

which supported 7 calves and 4 horses, strictly a hobby, and very poorly irrigated with free water. At the Board meeting the manager of this plant promised that if they couldn't irrigate without producing mosquitoes they would stop irrigating and this they did in the middle of August. He made one stipulation, that was that his neighbors' problems also be corrected.

With the exception of one grower, each of the 15 that have been cited into the Board has appeared. I think they have left feeling that they had a fair hearing and an opportunity to air any grievances they had to the Board. Now they expect us to clean up their neighbors' problems in the same manner.

Once a program of this type is started it must be pursued vigorously or the growers that have been cited and are being charged have a very legitimate complaint that they were singled out unjustly.

We do not intend to allow this to happen, we intend to continue this program through 1973 and/or until we find a more productive approach.

I have deliberately left the specifics of the legal part out because Don Murray will cover this very thoroughly after the recess.

## OBSTACLES PRESENTED BY THE STATE AND REGIONAL WATER RESOURCES CONTROL BOARDS

Dwight C. Baier

California Water Resources Control Board

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The California legislature in 1949 passed the Dickey Act creating state and regional water quality control boards and charging them with the responsibility of abating water pollution. For the first 20 years, the major effort of the boards was the control of pollution by industries and by cities. Personnel employed by the boards were generally civil engineers or sanitary engineers. An opinion by the Attorney General declared that the Dickey Act also applied to agricultural discharges, and the water quality control boards agreed to consider agricultural wastes after clarification of the problem.

The engineers recognize that city sewage wastes may add 300 parts per million (ppm) of minerals to water which already has a natural content of 500 ppm; the sewage discharge then would be 800 ppm. The philosophy developed by the engineers was to permit a necessary increment of mineral content but no more.

The Porter Cologne Act, the strictest water quality control act in the nation, was passed by the California legislature in 1970. The State Water Resources Control Board then addressed the agricultural waste water problem and employed me, the only agriculturist on the staff.

There are various opinions relating to the disposal of irrigation waste water. Water quality control would be improved if every farm in the State were in a well-managed drainage district with irrigation waste water conveyed to a proper place and treated in a proper manner. Since this could also benefit mosquito control, districts should encourage the formation of drainage districts. However, some drainage districts discharge water into the San Joaquin River and there is a question of how to establish discharge requirements for these districts.

When a farmer irrigates he puts water which contains natural salts onto his land. The plants use some of the

water, some evaporates, and the salts are left in the soil. With each irrigation the salt content increases, and eventually must be leached out if agriculture is to continue. If there is a subsurface drain, the excess water and salts move to some disposal point. The more efficient the irrigation, the saltier will be the discharge, perhaps up to 3,000–4,000 ppm. If a poor irrigator runs much more water than he should, he may dilute the salts so that the waste has only 1,000 ppm. If the Water Quality Control Board were to set a discharge requirement on a drainage district using the traditional approach of the engineers for city sewage plants, it could be demanding "use lots of water because you have to dilute the salts". This would be pushing the farmers towards inefficient irrigation.

Also, if farmers would be under discharge requirements in a drainage district, they would oppose forming such agencies. The Water Quality Control Board could provide surveillance over a few drainage districts but not over every individual farmer in the State. A way must be found to enable drainage districts to exist without having to pump good quality ground water to dilute the discharge, which would involve a serious waste of the resources of the State and expense. Rules for handling sewage discharges cannot apply to agricultural wastes.

The Porter Cologne Act adds to the duties of the water quality control boards the prevention of nuisances. If the waste discharge creates a nuisance, it is the duty of the boards to abate it. Legal council has confirmed that mosquito breeding caused by a waste discharge falls within the responsibility of the water quality control boards. Mosquito abatement district personnel are therefore encouraged to attend regional water quality control board meetings and to get acquainted with the personnel and the program.

## HOW COMPATIBLE ARE MOSQUITO CONTROL AND AGRICULTURE?

C. Brunel Christensen

Department of Agriculture

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Agriculture has a role in the control of many pests, including mosquitoes. The Department of Food and Agriculture has a detection force to note the presence and degree of infestation of the various pests. An analysis is made, then a decision on procedure: whether to attempt eradication or to accept a reasonable level of control. Each pest is treated as an individual problem. Consideration is given to biological control, physical control, chemical control, or to all together.

Newcastle disease in poultry is a current problem. The State Department of Food and Agriculture and the Federal Government believed eradication was desirable and feasible. After a year of effort, costing about \$30,000,000 and requiring the killing of 14,000,000 chickens, the disease has not been observed for the past 10 days and should be approaching eradication.

The pink bollworm in cotton came into California a few years ago from the southern states and is now out of control in the Imperial Valley. Control in the San Joaquin Valley is still effective and consists of several measures: about 1,000,000 sterile male moths are released per day, there is a plow down or no-host period, and some spray is used.

The Comstock mealy bug in citrus is in an eradication program requiring the use of many sprays and biological control. The woolly white fly, another citrus pest, came from Mexico and occurs primarily in the San Diego area. A quarantine line was established north of the infested area to try to contain it. Three years ago a spray program costing \$1,200,000 accomplished nothing except possibly to make matters worse by disrupting biological control. The citrus industry realizes that this pest eventually may spread throughout the citrus area of the State, but hopes to contain it as long as possible. Biological control with an intro-

duced parasitic wasp yields about 90% control in Florida and Mexico and is slowly building up in the San Diego area.

The Department has been criticized by some legislators and by the legislative analyst for its pesticide program. However, it is the best pesticide program in the nation and in the world. It is a four-pronged program developed to permit continuing use of pesticides:

1. Registration Program – Every pesticide, herbicide, and fertilizer is tested, analyzed, and reviewed for value and control use.
2. Licensee – All applicators, both ground and air, salesmen, and advisers are tested and licensed. Licensing and examination procedures are the best in the country and are being improved.
3. Health and Safety – Interdepartment cooperative regulations specify reentry time for the return of workers to a field which has been treated with insecticide.
4. Consumer Protection – All potentially affected foods are tested for residues. Tolerances for residues are reasonable and within federal tolerances. Large markets throughout the State are tested periodically. Water is also tested for residues.

Involvement of agriculture with mosquito control parallels involvement with other activities in the State. Previous efforts to achieve mosquito control by drainage were not sufficient in most areas. It is believed that it will take time to encourage the necessary changes and there is concern relative to who will pay for the changes. If they are of benefit to agriculture, then agriculture should be willing to pay for them. If they merely provide public benefit, then the public should support them.



## DISTRICT VIEWPOINTS ON USE OF THE LEGAL APPROACH

J. Warren Cook

Madera County Mosquito Abatement District

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The Madera Mosquito Abatement District is using what we feel to be a well rounded program.

First, we have a regular program of inspection and spraying of mosquito sources. To prove a point, we have cut down on our spray activity, due mainly to source elimination.

Secondly, we are involved in a year around public information program, as well as an educational program. Talks are given to service clubs, there are news articles, radio spots, etc., and we have compiled a book we call "Mosquito Research Unit Guide for Teachers". On request we furnish bookmarks, literature and a demonstration kit on mosquito life cycles. This is geared for all classes from the fourth grade to advanced biology since it permits teachers to use information needed for their particular classes.

Thirdly, we practice legal abatement action against consistent and flagrant offenders. Ours is a three-step system, perhaps four, if you count the many verbal approaches that are made. The system consists of a warning, a citation and, as a last resort, legal abatement action. It is described in the Proceedings of last year's Conference (Cook, 1972).

Under the topic we are discussing "District viewpoints on use of legal approach", I can give four examples of our use of the law to abate bad sources. In one case we followed the law exactly and served the City of Madera with a "Notice to Abate" a nuisance on their sewer farm. We listed improvements to be made, the time in which they had to make them and the date of their hearing, but left the door open for them to submit any alternate method of correcting the source they might have. This was served by the Sheriff on each council member and the City Administrator.

At the board hearing they submitted an alternate suggestion as to how they would like to correct the source, and it was accepted. The source was corrected, and it needed only to go as far as the hearing before the Mosquito Abatement District Board. The District spent at least twenty years trying to get the City to do something about the sewer farm, with no response, before we invoked the law.

The second case was against an individual, and we followed the same procedure as with the City. At the hearing in September, 1971, he agreed to submit plans engineered by the Soil Conservation Service. This was to be presented at the December 1971 Board meeting, but the S.C.S. did not have them ready. In January 1972 he sent a letter to S.C.S. authorizing them to supply the District with a copy of his proposal. This was done, and he was to appear at the January meeting to present them, which he did not do. He also failed to appear at the February meeting. At this meeting the County Counsel was instructed by the District Board to draw up and have served a "Notice to Show Cause". This was done, and the individual was given until the March Board Meeting to have the improvements made that he had

submitted. If not, he was to be at the meeting to show cause why it was not completed. He did not appear, and the Board instructed Counsel to proceed with the legal abatement action.

Section 2272 of the Health and Safety Code, states "The nuisance may be abated in any action or proceeding, or by any remedy, provided by law".

Our Board did not want to go onto this man's property and relevel, put in return systems, or anything else; they wanted him to do what was necessary and for him to accept his responsibility. With the Board's feeling in mind, the County Counsel made up and had served an "Order to Show Cause for Temporary Injunction". A Preliminary Injunction was granted by the Superior Court in Madera on July 26, 1972. This should have stopped all breeding, but instead he continued to water just as he had before. On August 23 we were back in court with a contempt charge against him.

We felt and still feel the case was very strong against him for contempt. The Road Department had issued a fine against him for flooding the road after the injunction was issued, not counting all the larva samples we had, and our having to spray up to twice a week on some of his parcels. On Friday, before we were to appear in court on Wednesday, the main defendant went to the hospital with cancer of the lung, and was given less than six months to live. This left his 23-year-old son, who is a partner, all of the responsibility. I cannot say it did, but with all the evidence we had, the sickness of the father must have influenced the court, which did not find him in contempt. We still have our injunction, and the Judge lectured the boy very hard on what he was toying with, and assured him the Mosquito Abatement District would win in the end.

The third case was against an abandoned potato shed located inside the City limits that had been left with all vats and ponds full of water and rotten potatoes. The owners were going through bankruptcy, and we had to serve notice on the Bank of America, City of Madera, Southern Pacific Railroad (landowners), as well as the owners. We worked with each and every one of these people from March of 1972 until August, with no action being taken by any of the parties, with the exception of many verbal promises.

The P G & E turned on the power for us in April so the owners could pump out the vats and ponds; but until the order to show cause was served and a hearing before the MAD Board in August, no physical action was taken.

This again went only as far as the Board hearing, where a certain time limit was set to have the necessary work done. It was taken care of and the source eliminated.

Just this past week the City Engineer called me and thanked me for getting them on the ball and bringing these ponds to their attention. It seems the City of Madera and S. P. are working out plans for the City to lease the ponds, install a lift pump, and use them for flood control.

The fourth case happened in October 1972 — the Sheriff served a temporary restraining order signed by the Superior Court Judge on an individual leasing approximately 280 acres from Southern Pacific Land Co. The notice was also served on S. P. Following service of the temporary restraining order, a hearing was to be held in Superior Court on November 3, 1972, but the parties stipulated to a preliminary injunction; and the necessary work was to be completed within 90 days. We have just given them another 90 days due to the wet weather we have had. The reason for the temporary restraining order is an attempt to speed up the proceedings.

We spent almost a year on the second case, and all this time the offender continued to do nothing to lessen the ponding of water and raising of mosquitoes, tuning out our pleas and threats.

We hope this new approach will speed up not only discussions, but action by the individual concerned, because

the restraining order will keep him from watering poorly leveled ground or misusing water so as to cause ponding.

From statements made to District employees, myself personally, and the Board of Trustees by members of the City Council, Board of Supervisors, and citizens of our County, I feel we have the backing of the majority in what we are doing. The local newspaper has told me many times that we have their 100% support, and anything they can do will be done — the local radio station has responded in the same way.

I think most people are sick and tired of the few causing problems for the many, and are glad to see us taking this approach.

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## VIEWPOINT ON USE OF THE LEGAL APPROACH TO MOSQUITO CONTROL

W. Donald Murray

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The Delta Mosquito Abatement District in 1972 did not have any insecticides which would provide effective control of the pasture mosquito, *Aedes nigromaculis*, with repeated applications over extensive acreages. Therefore, the growers who were producing these mosquitoes had to be told why their agricultural practices were producing mosquitoes, and what they must do to stop creating a public nuisance.

This District had been making person-to-person contacts with growers year after year. It had sent letters to growers for over 20 years, yet in most cases there has been no change whatsoever by the grower. Contacts, whether indirect or direct, without meaningful follow-up have not accomplished mosquito prevention. In 1972 the District took several steps to provide meaningful contacts and follow-up.

A formal policy letter was prepared and mailed to all growers who produced unacceptable numbers of pasture mosquitoes during 1971 and 1972. This was a strong letter, yet it invited cooperation. The mosquito producers were advised that they were producing unacceptable numbers of mosquitoes, and that steps would be taken by the District if they did not correct their practices. This policy letter stressed four points:

1. The deliberate and direct flooding of unlevelled or inadequately prepared land is strictly forbidden if it results in mosquito production.
2. Using adjacent unlevelled land for drainage from crop land is forbidden if it results in mosquito production.
3. The production of mosquitoes by any grower from land close enough to seriously affect populated areas is forbidden. The limit will be at least 2 miles, but can be

somewhat farther from the populated areas.

4. All irrigated alfalfa fields and pastures beyond the 2-mile limit must be put on a limited time table for correction.

The letter explained what action might follow if unacceptable mosquito production continued. This could be a citation, a board hearing and a charge for spraying, or it could be a referral to the District Attorney resulting in a fine for maintaining a public nuisance.

The letter was sent to the mosquito producers by certified mail, the red tag on the envelope helping to attract their attention. There has been much response and many corrections. Some of those who failed to make changes were given a citation and invited to a hearing before the District Board of Trustees. The citations were always delivered by two members of the District staff, not by mail or by a sheriff. Out of 15 citations during 1972, all but one grower came to the hearing and all meetings have been pleasant. Color slides of the specific problems are shown to each grower. The hearings have been rather low key; two main points have been stressed. One, the District staff will work closely with the grower, and two, if necessary the District will spray and charge the grower for the costs.

The growers do not like this legal approach; they would like to be left alone. But the District works for everyone, including about 100,000 people, and 400 or so major growers should not be permitted to destroy the quality of the environment for everyone. The primary reason for the existence of the District is to reduce mosquitoes to a tolerable level; wanton or careless degradation of the environment is no longer acceptable.

## RESULTS OF ABATEMENT HEARINGS

Theodore G. Raley

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The Consolidated Mosquito Abatement District has been able to cut its tax rate in half since it was formed in 1946; at that time it was \$.15, now it is \$.075. In 1947 a policy was adopted by the District Board stating that the person creating the mosquito source was responsible for its correction. The Board also took the position that the District would be responsible for correction and maintenance of natural mosquito sources.

The District did not rush in too quickly to confront agriculture with this policy. The best position appeared to be to first get a program organized and make the people aware of the District. There were annexations in progress so that by 1951 the size of the District had increased from about 250 square miles to a little over 1,000 square miles. During that time the District purchased equipment and tackled natural problems. A direct approach was made on the disposal of commercial liquid wastes, and with the help of the County Health Department, the County Counsel and the Fresno Mosquito Abatement District, the County Board of Supervisors was prevailed upon to pass an ordinance covering the disposal of industrial wastes in Fresno County. Since that time disposal of these wastes has not produced a significant problem. The districts also obtained an ordinance covering household waste disposal, and they worked directly with the cities on the improvement of community waste disposal sites.

The Board of Trustees of the District finally believed it was appropriate to direct full attention to the top priority problem, the control of mosquitoes related to irrigated agri-

culture. The first abatement hearing was held in 1955, when the District cited five property owners, involving about 2-½ square miles. This group was cited to the same hearing because the properties were adjacent and no one could be selected over the others. The problems were on more or less marginal land which prosperous ranchers were developing for later use. These growers brought a lawyer to the hearing. The District conceded that in order to leach out the alkali it was necessary for the grower to allow water to stand, although alternative procedures might have been available. The District took the position that the burden of mosquito control on these properties should not fall on the taxpayers. The lawyer withdrew from the case because he had been misinformed by his clients; the growers agreed to pay for a part of the work done by the District during the preceding year; and the land caused very little problem in succeeding years.

There have been other abatement hearings in other years. In each case the owner was given full opportunity to respond voluntarily before a hearing was called. In each case the response has been good. The Board has indicated willingness to bill the growers for work performed by the District, and to take whatever steps may be necessary if the problem continues to cause an excessive spray load for the District. The growers who have come to the hearings have made or are in the process of making corrections. During this past winter about 400 letters have been sent thanking people for the help they have given the District in the past year in reducing the need for spraying.

# EXPERIENCES IN THE PERSUASION OF PROPERTY OWNERS TO ALTER THEIR LAND OR WATER-USE PRACTICES

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With a large number of pastures (approximately 10,000 acres) that were raising resistant *Aedes* mosquitoes, the Sutter-Yuba Mosquito Abatement District commenced a program of source reduction in 1971.

Initially the District's plans were to survey the known "bad" pastures to determine the extent of the problem so that we might understand what it would require in the way of equipment and labor to correct the existing causes.

After the survey, we developed a priority list of pastures using degree of insecticide resistance, number of residences in the area, size of source in relation to cost of correction and other criteria.

As it sometimes happens, we didn't start on the property with the highest priority. This was due to an exceptional offer of cooperation by a landowner with a lower priority. We thought that Mr. Morrison raised mosquitoes due to poor water management, so we offered to irrigate for him; he accepted. One of our employees, who had experience in irrigation, took on the project. When the land was previously irrigated it would produce about thirty mosquitoes per pant leg; after we finished, it only averaged one per leg. We explained to Mr. Morrison how we irrigated. He then tried to duplicate our technique and he also was as successful. He continued to irrigate the rest of the growing season without producing enough mosquitoes to spray.

We have had people who were not too interested in our program. Mr. Ross lived next to a municipal golf course and had several hundred acres of pasture that were producing mosquitoes which were annually becoming harder to kill due to insecticidal resistance. We sent a letter to Mr. Ross outlining our concern of the level of resistance in his mosquitoes and offered to help him correct the problems that caused his operation of his property to raise mosquitoes. Several weeks after we sent the letter we made a follow-up visit to him and found that our letter had been thrown away upon receipt, rather than being carefully studied.

Mr. Ross had, several months earlier, leveled 120 acres of pasture to satisfy a need he felt was necessary. He did this without prompting from the District. At the time he had received our letter he had not recognized the value of his land leveling in regards to reduced mosquito production. By the end of the summer he must have noticed the differ-

ence in the properly leveled field as compared to the majority of his ranch, because he sought out the Source Reduction Foreman and asked for the assistance we had offered in our letter.

Not only have we had a good response from Mr. Ross, in that he has taken more awareness of what causes mosquitoes, but he has been going around the community telling people what he has done and that their places could be better, too. We know this is true because they have been asking for help -- "just like you did for Mr. Ross".

Not all of the landowners have been as cooperative as Morrison and Ross, and Mr. Cognina may yet live up to our "dreams".

Mr. Cognina farms 140 acres of pasture and due to land lay-out, irrigation practices, etc., he has raised 50-70 mosquitoes per leg with each irrigation. These mosquitoes do not stay on Cognina's property; they often move to the north. In 1971, we spent \$2,000 on Mr. Hoffman (northern neighbor) and \$850 on the Cognina property, on aircraft cost and insecticide. Because of the high expense on the adjacent property, during the winter of 1971-72 we sat down with Mr. Cognina; a BVC engineer and a biologist; an irrigation-pasture specialist from the University of California (Davis); and District personnel to discuss ways of controlling mosquitoes by land use changes. It was decided that Mr. Cognina would level and reseed part of the property, part would be untouched, and the remainder would have some new irrigation and drainage ditches dug to improve the potential for better irrigation practices; the latter was to be done by the District. In subsequent years the remainder of the land was to be leveled and reseeded. This was all to be done over a period of five years without Mr. Cognina removing any of his brood cows; the improvements to be paid for by improved pasturage at the end of ten years.

The District has completed its assigned task, but Mr. Cognina has not leveled any land, nor has he taken advantage of the work done by the District. The only "plus" value of Mr. Cognina's operation was that he did not irrigate the land that was scheduled to be leveled.

This has been only three of the six projects that we have carried to this degree of completion and the other three appear to have the same ratio of good cooperation vs. slow cooperation.

# LEGAL ACTIONS FOR ABATEMENT OF MOSQUITO BREEDING IN THE SOUTHEAST MOSQUITO ABATEMENT DISTRICT

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The Southeast Mosquito Abatement District since start of operation in 1952 has undertaken several actions up to and including legal action in court to obtain mosquito breeding corrections by mosquito producers. I will not detail the exact letters sent to the mosquito producers, but will summarize for a number of cases.

1. A dairyman who was a big producer of several species of mosquitoes in permanent pastures:
  - a. Over irrigation resulting in breeding of *Culex tarsalis* and *Aedes nigromaculis*.
  - b. Inadequate handling of dairy waste from milking barn, thereby causing production of *Culex quinquefasciatus* and *Culex peus*.
  - c. The first procedure was attempt of the field operator to get correction by verbal means and admonishment. Since this method did not work, a letter from the Manager was sent the dairyman advising him of the laws covering this violation as well as the procedure that would be followed if he did not correct the mosquito breeding. Since he did not correct the mosquito breeding, he was written a letter advising him to appear at a hearing at the next Board of Trustees' Meeting. Since he did not appear for the hearing, a case was filed against the dairyman on several counts which included the violation of several sections of Chapter 5 of the Health and Safety Code and nuisance provisions of the Los Angeles County Health Ordinance. This is not the procedure recommended by most district attorneys since a good procedure is supposed to be covered by one major provision of law. In any case, the Defense Attorney met with the Manager and Prosecuting Attorney and agreed to plead the dairyman guilty on one count if the other charges were dropped. This dairyman served as an example to the several hundred other dairymen in the District so that all that was needed to get corrections was a letter of warning.
2. Improper disposal of brewery wastes. One of the largest breweries in the District handles plant floor washings and other wastes in such a manner that heavy mosquito breeding was caused in several miles of flood control drainage channels. The species involved were *Culex quinquefasciatus* and *Culex peus*. A letter of warning and requirements were sent the management of the

brewery which resulted in a number of actions. The brewery called in the County Supervisor in whose District the brewery was located as well as other politicians. After suitable conferences, other agencies including the Los Angeles County Flood Control District, the County Department of Health Services, the Los Angeles City Public Works Department, and the Los Angeles County Engineer were called in resulting in a program of corrective action that will in the near future abate the problem.

3. Swimming pools. There are over 100,000 swimming pools in the District of which in the winter approximately 700 are neglected and breed mosquitoes one time or another. The rest of the year at any one time approximately 300 are found to breed mosquitoes. A number of these pools are located on properties which have various legal problems. The property may have been repossessed with the former owner still occupying the property. The property may be on the verge of repossession. The property may be owned by a person in a very difficult circumstance such as widow with 9 children. The procedure followed is to attempt to get correction verbally or by written notice by the field operator. If this procedure does not work, letters are sent to the property owner and occupant listing the problem and the legal aspects of the action. The difficulty in this type of abatement is to find out who the property owner is. Much time can be consumed in search of County records. We currently have 4 violators who refuse to correct the violation and notice of hearing. These are to be placed in the hands of private legal counsel to get the records searched so that the appropriate person or corporation can be charged for the violation.

In my opinion, the current laws covered in the Health and Safety Code from Section 2270 through 2289 can be used effectively by mosquito abatement districts to obtain abatement of violations. When it appears that real resistance is to be encountered from the violators, and by this I mean legally, then additional legal approaches should be considered and these include use of local county health department nuisance ordinances. If the mosquito breeding is in a city, the nuisance provisions of the city no doubt could be used. Furthermore, advantage can be taken of the nuisance provisions of the Health and Safety Code under the extraordinary powers of the Director of Public Health of the State of California.

## A PLANNED PUBLIC INFORMATION PROGRAM

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At Turlock Mosquito Abatement District, we feel that legal procedures to eliminate a man-made mosquito source should not be begun if they can be avoided. If it cannot be avoided, then we feel that general public support of the District in its efforts to enforce the law is vital as it is for any program a district may pursue.

In addition to determining mosquito sources, analyzing why each one exists and contacting property owners, we have embarked on a long-range public information program. The program is planned to inform the general public on the current mosquito problem and what the community and the District must do to cope with it. On April 1, 1971, we joined with three other districts to develop a planned public information program.

Turlock, East Side, Madera and Kings mosquito abatement districts signed a one-year contract with a public relations consultant for materials and services designed to assist the districts in their program of public information. The consultant supplied the districts with radio announcements, news releases, posters and consultation each month as listed in the schedule of services.

We prepared a basic public information outline to be used as a guide for selection of subjects or topics. The main headings were:

- I. Stating the problem;
- II. Background information on mosquitoes;
- III. General education on mosquitoes;
- IV. Solutions — mosquito prevention;
- V. Role of the Districts and the Community.

During the one-year term of the contract with the four districts, managers made contacts with newspaper reporters and editors, radio broadcasters, utility service people, librarians and anyone else they could think of that could disseminate information material to the public. When the contract expired in March, 1972, we felt we had gained enough experience to carry on without the services of a public relations consultant.

The four original districts, plus four more, joined together and formed the San Joaquin Valley Public Information Committee. This Committee held its first meeting at T.M.A.D. April 11, 1972.

We worked out a budget, hired a cartoonist and a copy writer and went on about the business of continuing our public information program.

This Public Information Committee meets once a month with our staff. We discuss and select two topics for each

month to be used as the theme for radio spots, news articles, bookmarks, and stuffers. This past season we have concentrated on materials for radio, daily or weekly newspapers, bookmarks for county library systems, and stuffers in monthly utility statements. We have done this because it has been proven the most economical means of disseminating information. Radio stations broadcast our announcements free of charge as a public service. Newspapers occasionally print our news releases as presented or will often call for a special article by their staff reporter. At Turlock, our local utility mails 31,000 statements each month and will include our stuffer at no cost to us. Our county library systems hand out 6,000 bookmarks at \$5.00/m. We could extend our public information efforts to the local schools, once-a-week shopping news, monthly trade magazines, free outdoor advertising (Outdoor Ad Association of America) and television. Our staff is also capable of writing and drawing material for pamphlets and leaflets.

The cost to each member district is approximately \$100 per year for development of written material and cartoons. This includes 12 bookmarks, 96 15-second radio spots, 24 news releases, 24 cartoons to be used on 12 bookmarks and 6 stuffers.

At Turlock, we feel the program has produced positive results. When District personnel contact property owners who produce mosquitoes, most of the contacts remember hearing something about mosquitoes on radio or reading about it in newspapers. When the county or city planning commissions hold hearings on proposed land uses involving topography changes, the public will often ask what effect the change would have on mosquito production. Not knowing exactly what effect it will have on mosquito production, the commissions will ask the District for recommendations. Several land use permits have been modified or denied in the Turlock area as a result of District recommendations. Cities, irrigation districts, County Public Works Department, and other public agencies are also becoming aware of their responsibility in comprehensive mosquito control.

The population in T.M.A.D. is estimated at 90,000. Through personal contacts with property owners and talks to service clubs and other groups, we estimate we were in contact with approximately 2,000 people. To be successful, a comprehensive mosquito control program must have the support and cooperation of the entire community. Our public information program is an effort to reach the other 88,000 people in our District.

The San Joaquin Valley Public Information Committee now has 13 districts participating. New members are welcome.

## STUDENT CONCERN WITH THE ENVIRONMENT

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In the 1950's and early 1960's an environmentalist was a person involved with environmental health. He knew a little about everything related to the environment. In the late 1960's when the word "environment" came into popular usage, the environmentalist became a person involved in impact studies of the ways various activities relate to the land. This is close to the fourth anniversary of the landmark date of the environmental movement in the United States, January 28, 1969, when the oil spill began at Santa Barbara. The people there were upset because one of the offshore oil platforms was leaking oil onto the beaches. That well is still leaking slightly, probably not much more than the natural leaks that have been occurring for thousands of years.

Students are concerned with the environment. Thirteen thousand well organized young people are looking for a mission and protecting the environment provides an ideal project for them. A few years ago there were riots at the Santa Barbara campus but there were not a lot of students involved in them. Perhaps 500 people were walking around the administration building but 13,000 were going to classes. Perhaps 5% of our students should not be there but should be out working.

Walter Hickel, formerly Secretary of the Interior, was under fire because of the oil spill. He was not recognized at the time as a leader in the environment movement. But in a famous letter to President Nixon he stated that youth in its protest must be heard.

Recently students decided that we should not be using 2-4-D on the campus. They approached the matter not from an emotional standpoint but with technical facts. They are a formidable group, well organized and intelligent. Earlier the students found that no one was listening, there was a lack of communication, and this helped to promote disorders. Then there always seems to be a few students who shake things up just to get attention.

Popular support for the environmental movement was accelerated when Rachel Carson published *Silent Spring*. In 1968 the DDT scare was rampant but mosquito districts had quit using it by that time anyway. Then came the oil spill and the concerns that public officials could not be trusted because some failed or refused to act.

Students face the future with alarm. They see pollution, urban sprawl, and animals becoming extinct; they feel that the science of ecology must be followed in order to survive. They believe that national and state leaders must be environmentally oriented and insist that they listen. Many recognize that we cannot stop development; a minority demand that society stop immediately, regardless of consequences. Some believe that rioting at Santa Barbara was related to the war in Indochina.

Students on the UC Santa Barbara campus have been quite involved in the field of ecology. During the oil crisis they actively participated in the bird and animal cleanup. They took part in a debate over a proposed freeway across the Goleta salt marsh. This salt marsh is a source of mosquitoes and the Goleta Valley Mosquito Abatement District would have appreciated having it eliminated. However, the students and some faculty opposed the freeway and wanted the marsh left alone. The freeway has been stopped; the environmentalists won!

Students formed a group called "Students for Environmental Defense", later called "Ecology Action". Most college campuses have similar organizations. On many occasions they have been a thorn to the administration, yet the demand for protection of the environment may be a good thing.

In the future students will take more interest in the environment. They are not fanatics; they are concerned about the future. The fact that most of these students will soon be voters must be considered by the politicians.

# TRUSTEES' CONCERN OVER LIMITATIONS OF PESTICIDE USE

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Orange County Mosquito Abatement District

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A trustee is appointed, selected, elected, or he manipulates his own appointment in some way. His capability varies depending on his environment and his interests. He has three basic responsibilities: to the public, to the employees of the district, and to himself. To be valuable, he must take an active part in his job as trustee. The ultimate responsibility for everything the district does rests upon the Board of Trustees.

Insects as a group are the most successful living things on earth, far outnumbering all other forms of plant and animal life. The insects, which the mosquito abatement districts strive to control, are a form of animal with jointed limbs, a skeleton on the outside, and three body regions: the head, thorax, and abdomen. They have three pairs of legs. Most insects have two pairs of wings but flies and a few others have just one pair and some have none.

Insects have been controlled more or less successfully with chemicals called insecticides. There are two major groups — stomach poisons for the chewing insects and contact poisons which are sprayed or dusted directly on the insects. The latter are usually used to control insects with piercing and sucking mouth parts.

Each trustee should ask, "What am I supposed to do about the use of insecticides?" Each trustee has a responsibility to the public and he should take it seriously. He should also ask, "What am I, as a trustee, supposed to do for the manager and the other people who work for the district?" and "How am I to tell them what to use and how to use it?" We employ expert personnel, they tell our board what they would like to do, and we give them authority. But do we always provide them the tools and assistance they require? We are not supposed to be technical experts; we are supposed to represent the people. We need help from experts who can properly advise our staff and our trustees.

Inasmuch as we are faced with the problem of a rapidly declining effective supply of "acceptable" insecticides, pesticides, larvicides, etc., I feel that our best approach is to design and implement a preventive program rather than a curative one. We in Orange County have gone a long way toward such a program by setting and maintaining a feasible and legal Source Reduction Policy.

The Orange County MAD basically is not an agricultural area, but one that is urban; we have 26 cities. Our problems, therefore, are appreciably different from those of districts in the Central Valley.

In practice we are not as harsh as it may sound from the statements which follow. The program is carried on by the District's manager and staff. They use a softsell, public relations approach with a huge stick behind them in the Health and Safety Code. Only in one case since the formation of the District have we come close to using the legal notification, and that proved to be a matter of failure in communi-

cations. Once the proper people were notified, the matter was resolved without resort to legal provisions.

The property owner responsibility clause and theme have worked exceptionally well in our area. We are fortunate in that we are a growing community and we are in on the very beginning of the preparation of the land, from natural to the forthcoming unnatural conditions. We are there early with the information that is necessary to prevent sources from occurring.

Here are the policies which our District has adopted:

**STATEMENT OF SOURCE REDUCTION POLICIES.—**  
**Definition of Source Reduction.—**The interpretation of the term "source reduction" is not limited to the physical act of reducing in size or eliminating a body of water, large or small, by the conventional methods of filling or draining. Source reduction methods in Orange County include any effort or act that will aid in the reduction or elimination of adult mosquito production from any body or container of water that is conducive to the breeding of mosquitoes.

These source reduction methods include the following practices by the person or agency responsible for the prevention of mosquito production.

1. Physical control by design and construction of new facilities and by draining or filling of existing facilities.
2. Biological control by use of mosquito fish.
3. Operational control by management and maintenance of agricultural irrigation and waste water disposal facilities in combination with weed control.
4. Chemical control by use of mosquito larvicides.

**SUMMARY OF BASIC POLICY.—**"Property Owner Responsibility" is the term that best describes the source reduction policy adopted by the Orange County MAD Board of Trustees. The District expects the legal owner of the property (Reference 2), including all public agencies (3), (4) and (7), to prevent mosquito production as the result of his operation or use of the land. The District does not charge the property owner for services rendered. The District does, however, furnish certain services free of charge, such as technical information, mosquito fish and emergency larviciding and adulticiding required to prevent a public nuisance resulting from an operational failure by the property owner. The District does furnish routine larviciding services to those public agencies (4) which operate street drainage and flood control facilities that are constructed and maintained according to good engineering practice. Legal abatement procedures are instigated when the property owner ignores his responsibility (5). Legal enforcement has never been required since a formal letter or an appearance before the Board has satisfied the property owner that the Board's source reduction policy is both necessary and equitable.



**AUTHORITY FOR POLICY.**—The laws relating to mosquito abatement districts and mosquito control are included with Division 3, Chapter 5 and 5.5, Sections 220-2426 inclusive of the California Health and Safety Code (1). The District powers are listed under Article 4. The specific powers which delegate to the Board of Trustees the authority to abate a nuisance are to be found in Sections 2270-2276.

**NEED FOR POLICY AND SUBSEQUENT SUCCESS.**—The source reduction policy referred to as "Property Owner Responsibility" was formally adopted by the Board of Trustees at their 92nd meeting, held September 17, 1954 (2). This policy was needed because the former policy for charging landowners for labor and materials did not result in source reduction or in the prevention of unnecessary mosquito breeding conditions. During the past 18 years, the property owner responsibility policy has been responsible for effective mosquito breeding prevention and control factors within the design, construction, operation and maintenance of major water use and disposal facilities as well as for the acceptance by the property owner, large or small, of his responsibility to prevent mosquito production on his property (8).

**LIST OF REFERENCES CONCERNING SOURCE  
REDUCTION POLICY FORMATION**

- (1) Laws Relating to Mosquito Abatement Districts and Mosquito Control. Excerpt from the California Health and Safety Code, 1971.
- (2) Private Property Owner Responsibility. Excerpts of the Minutes of the Board of Trustees from September 17, 1954 through March 4, 1955 in connection with the Irvine Company.
- (3) Public Agency Responsibility. Excerpts of the Minutes of the Board of Trustees from March 18, 1955 through April 14, 1955 in connection with the Orange County Water District.
- (4) Public Agency Responsibility. Excerpts of the Minutes of the Board of Trustees from September 21, 1956 through February 1, 1957 in connection with the Orange County Flood Control District.
- (5) Legal Approach to Mosquito Control in Orange County by Jack H. Kimball, Proceedings and Papers, California Mosquito Control Association, 1956.
- (6) An Educational Approach to Mosquito Abatement Via School Curriculum by Jack H. Kimball. Proceedings and Papers, California Mosquito Control Association, 1959.
- (7) Integration of Sewage Disposal and Mosquito Control in Orange County, California by Jack H. Kimball. California Vector Views, February, 1965.
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## PESTICIDES CAN BE COMPATIBLE WITH THE ENVIRONMENT

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According to Webster "compatible" is defined as existing together in harmony. "Environment" is the aggregate of all external conditions and influences affecting the life and development of an organism. A pesticide is not really compatible with the environment because even if the material is used only to kill a designated pest, such as the mosquito, that is part of the environment. In the purest sense, even an introduced biological control agent may be incompatible with the existing environment.

Man modifies his environment to suit his particular tastes; he will use any reasonable method to reduce or eliminate that which is objectionable to his tastes. To appraise the effect of an action on the environment, man may use a benefit-risk ratio. If the benefits are greater than the risks, the practice usually will be acceptable though not necessarily compatible. It is a basic precept with animal life that the environment is more or less hostile, so man strives to enhance environmental benefits and overcome environmental resistance.

One way to accomplish the objectives of managing the numbers of animal life is to alter the environment; animals that can specialize and adjust remain while those that do not become repressed or extinct. Another way is to change or modify the environment, which man has been able to do.

People have made statements about "destroying the environment". That is not possible; the environment cannot be destroyed but it can be altered or modified. Pesticides have modified the aquatic environment. There have been reports of large fish kills in the Rhine and Mississippi Rivers, accompanied by claims that pesticides were responsible. Closer examination then proved that pesticides were not involved in either case although millions of fish died. The federal water pollution control administration acknowledges that only 2.5% of the fish kills occurring in this country have been caused by pesticides; the principal source of pollution resulting in fish kills has been municipal sewage and industrial wastes. In California, a report of fish losses showed five cases in 1965, five in 1966, six in 1967, seven in 1968, ten in 1969, and none in 1970. These cases were all carefully investigated. Such losses are at a minimum in California and they can be prevented entirely with proper care in the use of chemicals. In 1963 there were eleven known cases involving fish losses amounting to millions of fish. The laws and regulations then in effect were not strong enough. All cases were of a minor nature except one, and all involved either illegal use or misuse; none resulted from proper use. The users of pesticides now generally realize that they must be more careful in their practices.

Pesticides can cause other effects on the environment. At Tule Lake, thousands of geese were lost because they ate zinc phosphide-treated rodent bait. A similar problem occurred in doves which ate strychnine-treated rodent bait. Grebes died as the result of a DDD treatment of Clear Lake to control the Clear Lake gnat. The pesticide was concentrated through the food chain of the fish, which were eaten

by the grebes. A similar case occurred at Big Bear Lake where the Department of Fish and Game used chemicals to control rough fish, and the pesticide affected some of the birds in the area.

These cases were the result of direct poisoning but there are other ways by which chemicals may affect the environment. The death of trout fry in New York resulted from DDT being passed from the mother to the egg, and when the tiny fish absorbed the yolk sack from the egg there was high mortality. In California, DDT had been mixed with rice seed before planting to control the first brood of mosquitoes in the rice field. Female pheasants ate the DDT-treated seed and the DDT was passed from the hen to the egg, causing injury and death of the young. This practice was carried on until 1962 when it was stopped and the problem disappeared.

There are subtle effects of these pesticides. Most insidious of these is the accumulation through the food chain — one organism containing a small amount of a stable or persistent chemical is eaten by another, and the material is concentrated through several steps of the food chain so that the top predators are exposed to large doses which may cause mortality. This was believed to be the cause of thin egg shells amongst the brown pelicans on Anacapa Island and the peregrine falcons.

Other alterations of the environment can affect wildlife, notably the elimination of habitat, food, and cover. Large-scale spraying of sagebrush in the northeastern part of the Great Basin region has seriously reduced the population of sage grouse. The herbicide used had no direct effect on the grouse but did reduce the food and cover. In the Central Valley of California unwanted vegetation along stream banks was eliminated to such an extent that there was a considerable reduction in the pheasant population. The ditch bank clearing could have been done by mechanical means and would have had the same effect on the pheasants but the herbicide was much cheaper.

Food sources of fish or wildlife may be depleted through the use of pesticides. Large-scale spraying of forests in Michigan resulted in a decline of the trout populations in the streams. The material used had not harmed the trout but the insect food source had been greatly reduced.

Pesticides may also harm predators or parasites of a pest species. After treatment the pest may rebound to much higher populations than originally existed. In another situation, the predators or parasites of a minor or secondary pest may be removed, allowing it to become a major pest.

With proper knowledge and consideration of the total effects of pesticides, they can be used safely and effectively. The benefit-risk equation should be considered in order to justify acceptance of a specific chemical at a particular time and place. Each material must be evaluated for each case if protection of the environment is to be achieved.

## FUTURE PESTICIDE METHODS WHEN CHEMICAL USES ARE CURTAILED

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Perhaps my topic should be "the new era". The program of the agency I represent is based upon knowledge of applied insect ecology. It was begun several years ago and the entomologists of the University of California provided much help. We believe that biological and natural control should be the backbone of any pest management system. I call our program "pest management based on biological control". It is not an eradication approach. For success it is necessary to understand the environment created by certain farming practices. To do so, the farmers will have to obtain the services of a professional entomologist. We want to encourage the farmer to buy the parasites raised since they will help the biological control. This means our pest management specialists must be problem solvers in order to integrate the cultural and biological controls and use chemicals only when necessary to guarantee the success of the field situations.

Problems must be solved on the site, on a particular field, or even on part of a field. There must be continuous monitoring to keep track of the populations. Sampling involves studying the various organisms involved in the habitat and their relationships. To exploit the biological and cultural controls we have to determine "what is a pest?"

There is a new publication called "Integrated Pest Management" available from the U. S. Government Printing Office at a cost of 55 cents. It states that the three main components of a pest management program are; (1) maximizing existing natural controls, predominantly by cultural methods to prevent the buildup of pests, (2) monitoring the concentration of pests and natural control factors present to determine the need for further measures, and (3) using the most appropriate techniques or combinations of pest suppression techniques only when necessary to prevent economic damage to the crop.

Flies may be used to illustrate our philosophy of pest management based on biological controls. We can tolerate a few flies on a chicken ranch unless there is a lot of fly-transmitted disease because then they are normally only a nuisance factor. The high cost of killing the last fly becomes a problem when resistance to pesticides accentuate the problems. Even though resistance to most chemicals is now apparent in fly control, many pesticides are still good as a bait. Bait stations are very useful. Most of the predators of dung-inhabiting flies are associated with decomposing material such as manures, and the parasites and predators are attracted to decomposing manure. Chemicals should be directed to the adult in a way that they do not interfere with the natural enemies in the manure. This poses the question "can we tolerate manure on the property?". A frequent cleanout program is a good way to control flies but high costs may prohibit this approach. Or we may try to provide a habitat for a natural enemy complex that will live in the manure and eat the fly eggs every day, and thus give good biological control.

Various predators eat the fly eggs and tiny larvae and there are some parasitic wasps which lay eggs in the late larvae and large pupae. Most of the beneficial insects work at night and are hidden in the day time. The natural enemy complex can control flies, though control is rarely perfect. Management is the key to success. Programs in progress for the past five years using integrated pest management techniques that augment the biological control program have documented the success of our management program. Pesticides can still be used intelligently if there are too many flies, but one should use pesticides only when one has to, and then "selectively" so as to restore the natural balance instead of replacing it with repeated chemical applications that always lead to "resistance" and ultimate failure of the pesticides! Integrated pest management solves these problems.

## ENVIRONMENTAL APPROACHES TO VECTOR CONTROL

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If a control program is to be employed involving chemical and biological technology, it should be referenced to the environment. The greatest hazard to a mosquito is dehydration, whether in larval, pupal or adult stage. Emergent aquatic vegetation is a vital part of the habitat of most mosquitoes. Although all mosquitoes do emerge from water, actually they come out of only about 1% of the available water, because open water space is hazardous due to predatory animals and the impact of wave action. A clean shoreline will result in the drowning of the young larvae. Management of water, then, requires a clean shore line. In the final analysis, what I am talking about is land and water management for mosquito prevention.

Mosquito source reduction has been used as almost a synonym of physical control of mosquitoes. Also, if the appropriate parasite or predators exist in a mosquito habitat so they can be made to function effectively, then mosquito production is prevented. Above all, none of the three technologies should be downgraded — the physical approach, the biological approach, and the selective use of chemicals.

A fundamental consideration of the physical program is to assure that water will run downhill; in other words, drainage must be provided. Water management has become complicated, however, because when water is transported by drainage, impurities which may pollute another area must be given consideration. Surface drains were the primary approach during the reclamation era of mosquito control. Salt marsh drainage illustrates this approach. Subsurface drainage, usually by tile, tends to lower the top water in the soil and keep the plant root level free of too much moisture. A third method is vertical drainage, which requires a breakthrough of the soil so that water can penetrate into lower levels.

Stream mosquito control may be carried out by minor drainage of the ponds in a stream or river bottom, so that flow can be facilitated. Then a small dam may be constructed to permit stream flushing to wash any larvae away.

Excess vegetation around a home does not produce mosquitoes, but it does provide a moist environment which protects the body moisture of adult mosquitoes. Cutting will reduce the harborage area for them.

One of the biggest problems in California is the irrigated pasture. After some degree of levelling a farmer plants forage plants which provide feed for livestock. Then he may provide hopeful management without adequate thought — he believes that the pasture will be good indefinitely and will never have to be modified — it is a "permanent" pasture. Livestock are put on the field, and settling occurs, and the animals compact the depressions. I must state: the field ages, there is no such thing as a "permanent" pasture. How can that field be rehabilitated after it has

been allowed to become sick? Perhaps 60% of the field will produce good forage, but 40% is sick and produces mosquitoes. This suggests an economic corollary — if 40% is in weeds of low forage value, then the farmer is getting only 60% efficiency. Therefore something should be done if only to solve an economic problem.

It has been noted that the Rural Environmental Assistance Program (REAP) has been dropped, which is unfortunate for California farmers and mosquito districts. The CMCA should stress to legislators and governmental officials the importance of federal funds such as these for purposes of agricultural rehabilitation. There are over 1,000,000 acres of pasture land in California. Conservatively estimated, one half of these acres are producing mosquitoes in significant amounts, and about one fourth produce both the pasture and the encephalitis mosquitoes, which are resistant to virtually every insecticide that is available to the control agencies. The problem cannot be solved by the use of insecticides, if indeed they continue to be available. It must be solved by environmental techniques, by land and water management.

About one year ago the mosquito control agencies were invited to contribute funds to the State Department of Public Health in order to make it possible to engage in demonstration projects, to point out ways of solving the problem. The Department acquired four registered engineers who engaged in these projects, presently about 70 in number. Designs have been developed for remedy. The problem, unfortunately is in getting execution of the demonstration within the engineering requirements. The place of engineering, or environmental management, in the control programs must be taken seriously. The year 1973 is high in precipitation. Dependence on continued use of pesticides is no longer sound. Our agencies must be versatile enough to prescribe the treatment needed for the mosquito source, whether physical, biological or chemical or a combination of these. There may be more than one way to deal with any individual source. Certainly the agencies must recognize that they can no longer simply superimpose an insecticide over the environment.

The day of employing laborers — the man off the street, the unemployed individual — is over! The man who handles these environmental solutions of tomorrow will have to be skillfully prepared to carry out a balanced approach of mosquito prevention aided by biological control technology and discrete use of chemicals.

There are many other water management problems. Culverts under highways must be installed to permit drainage. Catch basins generally have a container which holds water and is ideal for mosquito production. Back yard containers are subject to physical management. The Bureau of Vector Control has had input into large water impoundment structures since 1958. Had this not been true, many of the

water projects developed in California would be more troublesome than they are today. To date, unfortunately, no agency is willing to accept responsibility for a water removal system installed at the time of water delivery. The Environmental Protection Agency (EPA) and the State Water Quality Control Boards are now establishing more rigid requirements on removal of waste water, this subject is getting more attention. It is pointing more than ever to return flow systems. Water problems will have to be solved

on each premises.

Mosquito Abatement District Boards and their staffs should recognize the importance of thinking comprehensively about the prevention and technological remedies for mosquito control in the future. Some practices of the past must be changed. An environmental approach in its total connotation must be used. Staffs must be of sufficient quality that this modern approach can be realized, and that technology can be used scientifically and effectively.

## RESPONSIBILITIES AND SERVICES TO MOSQUITO ABATEMENT DISTRICTS

Boysie E. Day

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There are 31 talks to be presented at this conference by the staff and students of the University of California. This fact demonstrates more eloquently than I can express in words the level of commitment of the University. The University has an increased and intensified commitment to serve the public by providing information to the districts which are the action agencies. The University has one business only — knowledge. New knowledge is created by research and the University is the custodian of knowledge developed around the world. This knowledge is then imparted to others through the teaching of undergraduates, teaching of graduate students, and on-the-job training and research technology. This includes presentations to groups such as the CMCA, and active extension into the field to people who have a need and a use for the information.

When knowledge passes into the implementation stage a strenuous effort is made by the University to get out of the picture and let the action agencies apply the information. The University of California has the responsibility to conduct basic and applied research toward the development and application of practical and environmentally acceptable mosquito control strategies, and to make these findings available to the MAD's and to all other agencies concerned in this important problem. In response to the appearance of massive and widespread insecticide resistance, and in spite of a declining budget, the University's research effort has been expanded, extended and diversified in response to this serious public problem. The University has appreciated the support and confidence of the MAD's.

The University is conducting studies on the biology, ecology, population dynamics, breeding, feeding habits, dispersal, survival, and reproduction of mosquitoes. This is the basic biological research that undergirds the control programs of the districts and the University's applied research programs. The applied programs cannot function well in a practical system unless there is understanding of how the system works. Some work can be done with only a smattering of basic knowledge but a good program cannot be designed without knowledge of the fundamentals.

There is important research going on by the University in chemical control. The picture is now looking better than it has for the past three to five depressing years. There are exciting developments in the growth-regulating chemicals and other compounds involved in regulation of the life cycle of these mosquitoes. There is important research on larvicides and pupicides and adulticides. The genetics of resistance in the gene pools of these insects is an important study. Far out on the horizon there are theoretical considerations which may lead to advanced management of genetic systems of whole populations, offering promise for future sophisticated controls.

Much work is being done on the environmental impact and social impact of many of the mosquito control activities. A review of research programs throughout the nation and the world indicates that the University of California leads the world in this regard.

## STATE AGENCIES CONCERNED WITH MOSQUITO CONTROL

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In the Department of Fish and Game we have the responsibility for protecting and conserving the fish and wildlife of this state so that the people can enjoy them now and in the future. In carrying out this responsibility we try to achieve these objectives: (1) to maintain all species of fish and wildlife for their intrinsic and ecological values as well as for their direct benefits to man; (2) to provide for diversified recreational use of fish and wildlife; (3) to provide for an economic contribution of fish and wildlife in the best interests of the people of the state; and, (4) to provide for scientific and educational use of fish and wildlife.

The key to preservation of fish or wildlife is habitat or suitable food, water and cover. Birds, fish and animals, like man, must have a suitable environment to live in or they will die. With California's ever-increasing human population the competition for land is also increasing, and so we see wildlife habitat being converted to industrial sites, urban developments and agriculture. Without a doubt one of our biggest jobs is to preserve the remaining habitat.

Most wildlife habitat does not produce many mosquitoes. Upland type that produces quail, chukar partridge, doves and big game like deer, seldom creates mosquito problems. On the other hand, wetland habitat, or marshes and ponded areas, provide homes for waterfowl and other water-associated wildlife, but it can also produce hordes of mosquitoes. It so happens that wetland habitat is also in short supply, and we are trying our best to preserve what little remains.

Why is this wetlands habitat so important? It is important because it provides winter homes for most of the waterfowl that use the Pacific Flyway. It also provides food and living space for shorebirds, herons, egrets, grebes, rails, gulls and countless songbirds -- some of them rare or endangered species.

Each fall about 10 million ducks come to California. Their exact number depends on the success of the nesting season in Canada and Alaska. During periods of high production like 1956-58 as many as 15 million ducks entered the state. When drought periodically appears on the Canadian prairies and reduces the nesting habitat, production falls and California receives only about 7½ to 8 million ducks. In addition to the ducks, about one million geese migrate to the California wintering grounds.

Although California is primarily a wintering area some birds do remain in the state to nest. Each summer the adult and young birds total about 300,000 ducks and 25,000 Canada geese. Much of the duck nesting and all of the Canada goose nesting takes place in the Great Basin area of the northeastern part of the state. Another important nesting area is the Central Valley where "local" mallards make up most of the nesting birds. In fact, the majority of the mallards that are taken by California hunters are locally produced birds.

All of the waterfowl coming to California are seeking suitable habitat, which will carry them through the winter.

It has been estimated that at one time there were 5 million acres of wetlands in the state. The first definite record of the amount of wetlands comes from a land inventory that was made in 1906 by the U. S. Department of Agriculture. It showed that there were 3,420,000 acres of wetlands in the state. A similar survey in 1922 showed that under the economic pressures of agriculture, industry, and urbanization these wetlands had dwindled to 1,179,000 acres. By 1954 the wetlands survey of the U. S. Fish and Wildlife Service showed that wetlands had been further reduced to 559,000 acres. Since then additional wetlands have disappeared and there are now less than ½ million acres remaining. Of this remaining habitat, about 225,000 acres are in privately-owned duck clubs, while most of the balance is on the state and federal waterfowl areas.

These state and federal areas were acquired to preserve wetlands. The U. S. Fish and Wildlife Service has 12 national wildlife refuges in California that total about 200,000 acres. Our Department manages 8 areas that total over 50,000 acres. A joint agreement between the two agencies provides for the management of the areas with the following three major objectives:

1. To provide suitable habitat and living space, so that an adequate population of waterfowl will return to the northern nesting grounds in good breeding condition.
2. To provide adequate food to keep waterfowl from depredating agricultural crops.
3. To provide public hunting for as many hunters as possible.

The last objective has been expanded to include other recreational use such as fishing, birdwatching and sightseeing.

When we first acquired some of our areas we were not too concerned about the mosquitoes that we might produce. Some of the areas were rather remote from habitation, and only our personnel living on the area were bothered by the mosquitoes. But in time the urban sprawl moved out to reach our areas and it was not long before we were accused of being the source of all the discomfort that mosquitoes can cause. More recreational use of our areas has also brought a demand for more mosquito control. At one time most of our public use came from hunters during the fall and winter months. Now other uses of the area surpass the hunting use and we find that we are in the recreation business the year around.

Now as we develop our areas to make them more productive for wildlife we also make provisions for mosquito control. Fields and ponds are constructed in a way to have good water control. With proper water management we can help control mosquitoes and grow better crops of waterfowl food plants. Many of our cultural methods for growing food plants are dependent on water control. The main thing is that as we use water to improve wildlife habitat we do not

produce a large crop of mosquitoes. Researchers have found that many plants that are good producers of wildlife foods also produce very few mosquitoes. Most of these plants are of the emergent type with naked stems and include the spikerushes, alkali bulrush, and also some of the pond weeds. On the other hand, there are many plants that encourage mosquito production and produce little or no food for wildlife. These are such plants as cattails, water hyacinths, water primrose and the milfoils. Both mosquito abatement personnel and wildlife managers consider these plants as pests.

The Department continues to view with great interest the efforts being made to use fish in the control of mosquitoes. It is encouraging to learn of the value some of you place in the role of fish in your control programs. We hope that solutions to some of the resistance problems may lie in the expanded use of these animals in integrated programs. As you may remember through your Wildlife Committee our Department participated in a cooperative program a few years ago to identify fish of potential value for mosquito control purposes. These fish were collected by mosquito abatement districts throughout the state.

One of our areas is helping out in the production of *Gambusia*, these fish can control mosquitoes, but many times there are not enough fish to handle the problem. This is especially true in the spring when fish populations have been reduced by winter-kill. To help carry fish over the winter we have made available to the Sutter-Yuba Mosquito Abatement District a series of ponds at our Gray Lodge Wildlife Area. In these ponds fish can be over-wintered and then distributed in the spring to potential mosquito production areas. If additional efforts would result in the more effective use of this type of mosquito predator we would be glad to work with you in a cooperative effort.

We also make provisions for emergency treatment of mosquitoes on our areas, where it is evident that chemical treatment might be necessary. Each area budgets funds and service agreements are made with local abatement districts. In this manner we are ready for immediate action should treatment be necessary. Many people think that we are opposed to chemical treatment. However, the Department is very interested in working with the mosquito abatement districts in regards to their use of insecticides. We recognize that by encouraging the districts to help out in preserving marsh areas for wildlife we are in a sense advocating the use of chemicals rather than source reduction. We are also concerned about the hazard to fish and wildlife that can result from the use of insecticides. It is quite obvious, therefore, that we have an obligation to assist you in developing insecticide usages that are effective, yet not harmful to fish and wildlife.

Our major role in this regard is to evaluate the side effects on wild animals of new control chemicals or techniques for mosquito control before they become operational. In this way we hope that by the time the control procedures have been approved for the district's use the chemicals can be applied as directed without unwarranted side effects. In fact the intent is to incorporate a rather sizable safety factor into the recommendation to allow some leeway in applications before adverse effects can occur.

However, as the problem of insect resistance increases there has been a tendency to seek solutions to this problem by increasing application rates of available insecticides or through the development of new materials. Newer insecticides, like Dursban® for example, are highly toxic to both mosquitoes and fish at low levels. If new highly toxic materials are to be used, or if the application rates of insecticides in current use are increased the safety factor that provides protection to fish and wildlife will be reduced. This means that chances of unwanted side effects occurring will be greater and that additional care in the selection and use of insecticides will be required if losses of nontarget animals are to be avoided. We believe that close cooperation between conservation agencies and those agencies involved in mosquito control is required to prevent these problems from actually developing.

To that end we work directly with chemical companies, the Bureau of Vector Control and Solid Waste Management and your abatement districts in developing use practices for new chemicals. A prime example of our efforts in this area was a recent study of light oils used as larvicides. This study was conducted on one of our wildlife areas, and it showed that these light oils did not harm fish or wildlife.

We have also worked with your Wildlife Committee to inform the abatement districts of the geographical location of those species of wildlife that have been listed as rare or endangered. A copy of our publication "At The Crossroads" was distributed to each district. With this as a reference a district will be aware of these species and can avoid using control measures that might cause harmful secondary effects to them or their habitat. If more detailed information is needed please call on us. We prefer to be in on the early planning, than to be called in after the control measures have been completed.

Besides managing our own lands we are also interested in preserving habitat on private lands. The value of private duck clubs in helping to preserve the state's little remaining wetlands cannot be overlooked. In all, there are about 1,000 clubs and they control about 300,000 acres of land. However, these lands are not thoroughly developed so that only about 225,000 acres can be considered as wetlands. One way to compensate for the loss of habitat is by increasing the carrying capacity of the remaining wetlands. Many of the duck clubs are only partially developed, but by improving their holdings the clubs will be able to carry more birds. The Department of Fish and Game technicians and the soil conservation districts encourage duck clubs to improve their habitat by proper water manipulation, raising aquatic foods, and controlling the undesirable cattails and tules. As we do on our own lands we try to have clubs use management practices that will control mosquitoes. In our advice to them on how to improve their habitat we stress the importance of developing land so that it will not create mosquito problems. We also urge them to work with their local abatement district, especially if there are any problem areas. If a problem arises we want the district to get in touch with our local field personnel. Through a series of regional meetings the Wildlife Management - Mosquito Suppression Coordination Committee has been getting mosquito control and wildlife people together so that they can get acquainted and resolve potential differences before they arise.



There are also times when we have to call on you for help, mosquito abatement districts like to see the waterfowl hunting season start as late in the fall as possible. In that way the weather will be cooler when the duck club lands are flooded and fewer mosquitoes will be produced. Our Department also likes to see the season start late so that the birds are not driven into the rice fields and cause depredations to the crop before it can be harvested. For most areas of the state a later season usually means better hunting as the migration progressively increases until all the birds are here for the winter. Last year some hunters were asking our Commission to adopt the earliest possible season.

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However, thanks to the help of your Association we were able to convince the Commission to take the latest possible season.

In the future as water is distributed to new areas of the state by the California Water Plan we are hopeful that water will be available for improving wildlife habitat. It is also apparent that with the wider distribution of water the problems of insect resistance and urban sprawl will combine to make the job of mosquito abatement even more difficult. However, we believe that by continuing to work with you control programs can be developed that will provide effective mosquito control without harming wildlife.

# FISH AND NEMATODES – CURRENT STATUS OF MOSQUITO CONTROL TECHNIQUES

James B. Hoy<sup>1</sup> and James J. Petersen<sup>2</sup>

**INTRODUCTION.**—The topics that we've been asked to discuss today are quite diverse, in that they have in common only that they are both forms of biological control of mosquitoes. Nematodes, as biological control agents of mosquitoes, are currently under study by a relative few researchers and have not been used for mosquito control by any agency. In contrast, fish, especially *Gambusia affinis* (Baird and Girard), have been used for mosquito control for more than 65 years and have been studied extensively by both ichthyologists and those researchers directly involved in mosquito control. But, the most striking contrast between these two types of biological control is that one has the techniques for mass-rearing the agent well worked out, including reasonable cost estimates; the other is at the stage where we still must rely on natural or fortuitous stock.

For each control agent mentioned we must ask three basic questions:

1. Will the agent work? Implicit in the question is an inquiry about the results of field tests.
2. What are the side effects of the agent's use? (Will it become a pest or inhibit the success of other control methods?)
3. What is the dollar cost, that is, the direct cost of acquiring, distributing, and monitoring the effect of the agent? This cost should not be lumped with costs that are generated by side effects.

**PARASITIC NEMATODES.**—Parasitic nematodes are detrimental to their insect hosts in several ways. For example, the indirect "mode of action" of *Neoaplectana* spp. is interesting in that the nematodes carry a microbial agent which causes the death of a wide range of insect hosts. The direct "mode of action" of the more host-specific mermithid nematodes has a direct physical effect on the host similar to that of a small can opener; it punctures the host from the inside as it departs the host preliminary to the final stage of the worm's development. [See Poinar (1971) for a full discussion of insect control through the use of nematodes.]

During 1972, our attempts to use *Reesimermis nielsenii* Tsai and Grundmann against *Aedes nigromaculis* (Ludlow) had highly varied results. The best rate of invasion (and all invasions result in the death of the host) was 57 percent. That degree of success occurred when the preparasitic nematodes were applied at a rate of 10,000 per square yard. However, these tests were made in small plots and under varied irrigation patterns, which may have accounted for the great variation. The inundative releases obviously did not

achieve immediate control of the pest, but the worms may become established and eventually reach numbers adequate for control of the host mosquito. Our final results will not be determined until well into 1973.

In other tests with *R. nielsenii* (Petersen et al. 1972), the nematode was quite effective (80-85%) against *Anopheles freeborni* Aitken when it was applied at a rate of only 1,000 nematodes per square yard in a rice field. Also, a second generation of *R. nielsenii* was under way in that study area in less than 5 weeks. That rapid cycle suggests that inoculative releases of the worms might be effective, perhaps even against *A. nigromaculis*.

*R. nielsenii* is the most thoroughly studied mermithid known to parasitize mosquitoes. But, Drs. Poinar and R. D. Sanders of U. C. Berkeley are presently describing and studying the biology of a native mermithid that they discovered in *A. sierrensis* (Ludlow). The fact that this new species parasitizes an *Aedes* species in nature is especially encouraging because it may prove to be an effective control agent for *Aedes* mosquitoes.

The prospects for the use of mermithid nematodes as a group for mosquito control seem good. We already have one species that worked well on *Anopheles* in field tests. The undescribed species from *A. sierrensis* holds some promise. Additional species of mermithids have been found and await study and/or proper taxonomic description.

Beyond having good candidate species, the techniques of mass rearing *R. nielsenii* in vivo have been well worked out (Petersen 1973, Petersen and Willis 1970, 1972). During 1972 these methods were successfully applied at the Sutter-Yuba MAD facilities, on a scale four times greater than had been previously attempted. Furthermore, in vitro rearing is attempted in at least two laboratories, and growth has been observed (Sanders, personal communication).

In summary, during the last few years, much progress toward the use of parasitic nematodes for mosquito control has been made both in field studies and in the laboratory.

**FISH.**—Over the years, many species of fish have been nominated as candidates for biological control of mosquitoes: *Gambusia affinis* (Baird and Girard), *Tilapia* spp. and the white amur (*Ctenopharyngodon idella* Val.) are diverse examples. Nevertheless, in no case have the questions of efficacy, side-effects, and direct cost been fully answered. Even for the best studied species, *G. affinis*, direct cost is poorly known though efficacy is reasonably well established and rough estimates of the side-effects can be made.

The reason that the cost of *G. affinis* is poorly known is that we are still dependent on fortuitous sources of fish, and therefore the basic cost of fish cannot be established. Sewer ponds that are not heavily loaded with organic material (and that are not subject to slugs of toxic chemicals) are presently the main source of fish. Farm ponds, ditches, and bait dealers are occasionally sources. None of these sources are very reliable.

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Interest in *Gambusia* has increased significantly in the last few years. And we believe that there is a consensus that the limited supply of fish is the major problem that we face. This problem is under attack in several ways, i.e., development of sewer ponds as sources, culture of stock in managed ponds, and hatchery rearing. The Bureau of Vector Control and Solid Waste Management of the State Department of Public Health is studying the factors influencing the fish production of sewer ponds and hopes to have E.P.A. funding to enlarge the study soon (D. J. Womeldorf, personal communication). Staff members of Sutter-Yuba MAD, Northern San Joaquin County MAD and Fresno Westside MAD are in various stages of investigation of managed ponds as fish sources. The first two of these districts now have full-time fisheries biologists on the staff. Hatchery rearing (also referred to as intensive culture) of *Gambusia* is presently under study by personnel of Kern MAD, Orange County MAD, and the Western Insects Affecting Man and Animals Laboratory of the USDA.

All of these culture methods are in the early stage of development. Perhaps a year from now we will be hearing

submitted papers dealing with such topics as feed conversion rates, yield per acre, and how to write an environmental impact statement for a fish hatchery.

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## MOSQUITO REPELLENTS

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The initial successes with DDT ushered in the rise of this insecticide as an ultimate weapon for exterminating mosquitoes but soon mosquitoes themselves aided its downfall by becoming resistant to it. The story was repeated with other chlorinated hydrocarbons, carbamates, and organophosphates. Recent ecological awareness placed another damper on the chemical control of mosquitoes. The avenues to control mosquitoes then proliferated—pheromones were investigated, sterility methods are being developed, biological control methods are being extended and reevaluated, land and water are better managed and recently much effort is being made in the development of juvenile hormones or growth retardants. Combinations of these methods are now applied to control mosquitoes under the term "integrated control".

Though the value of these methods cannot be underestimated, it does not seem mosquitoes can be exterminated

completely and man may always have to contend with residual mosquito populations. The only way to cope with them would be with the use of mosquito repellents. We are in collaboration with Stanford Research Institute in the development of new insect repellents and in potentiating the existing ones. The SRI group has synthesized on the basis of structure-function correlations compounds of which three have proved to be superior or equal to the present standard repellent "deet" (diethyl toluamide). In this laboratory we have succeeded in enhancing the protection time of existing mosquito repellents by two to three times with the use of fixatives. We have also succeeded in enhancing the abrasion resistance of existing repellents and their water washability 14-15 times with the use of synthetic binders.

Having a cosmetically acceptable, abrasion resistant, low cost 24 hours repellent now seems a distinct possibility.

# AERIAL APPLICATION OF MOSQUITO ADULTICIDES IN IRRIGATED PASTURES

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Larvicidal programs have been the preferred method of control aimed at *Aedes* mosquitoes in irrigated pastures in California. Due to the appearance of high level of resistance in pasture and other mosquitoes to most of the currently used larvicides, efforts have been recently made to develop adulticidal measures and to evaluate application techniques and adulticidal materials. Akesson et al. (1969) presented information on the efficacy of ULV aerial application of chlorpyrifos (Dursban®) in rice fields. Mulla et al. (1972) evaluated several quick knockdown adulticides against pasture *Aedes* using low pressure delivery system mounted on a Call Air aircraft. Results of these trials were erratic, although moderate to high level reduction in adult activity was achieved in some of the treatments. The nozzles and pressure system utilized in these experiments yielded coarse droplets.

During the 1972 trials, however, it was deemed desirable to develop a system that will generate a droplet spectrum in the range of 20-40 microns. To achieve this end, two propeller driven Hypro pumps were installed on an aircraft and a boom carrying 9 (25-TX-12 Spraying System) nozzles was affixed to the aircraft. This system was tested under laboratory conditions and found to develop 450 psi pressure using some commonly available petroleum hydrocarbons. This system was further modified by installing an electric pump to provide for more dependable operation, and was evaluated for efficiency using several quick knockdown insecticides.

## Methods and Materials

**EQUIPMENT AND MATERIALS.**—A Call Air A-9 plane of the Kern Mosquito Abatement District was fitted with a high pressure delivery system. The pressure initially was obtained by means of two Hypro propeller driven pumps, but later these pumps were replaced by an electrically driven Pesco hydraulic propeller feathering pump (Model 300). The boom was supplied with 9 (25-TX-12) nozzles, but it was found that no more than 4-6 nozzles were needed for ULV applications of 6-8 oz/acre.

Although this high pressure delivery system was tested in the laboratory and found to develop a pressure of 450 psi, the system even after installation of the electric pump was quite erratic developing variable pressures ranging from 100-75 psi, depending somewhat on the viscosity of the materials sprayed.

The altitude of the aircraft during flight was held between 20-50 ft in most experiments. In one of two tests the altitude was held at 100-150 ft. The swath widths were predetermined by flying the aircraft at certain lines. Swath widths reported for the various tests are only theoretical and not effective swath widths.

**SAMPLING.**—Assessment of the efficacy of adulticidal measures applied to small experimental areas is extremely difficult. One method of assessment commonly employed in studies on adulticidal measures is that of placing caged mosquitoes in treated and untreated areas. This technique yields valuable information on certain aspects of the treatment such as deposit pattern, drift, relative efficacy, etc. For example, Gillies et al. (1972) have used this technique to good advantage for gathering information on the susceptibility of mosquitoes to adulticides applied by nonthermal aerosol generators.

Placement of caged mosquitoes, however, does not provide for an accurate inference with regard to the natural populations of mosquitoes, especially *Aedes* in irrigated pastures. Caged mosquitoes are directly exposed in the path of a moving aerosol cloud generated from ground or aerial equipment. The height at which these cages are set will influence mortality of mosquitoes exposed in cages. Those placed near the ground will probably suffer less mortality than those placed higher up on stakes. In nature, populations of *Aedes* in pastures when not actively flying were observed to rest in cracks in the ground and at the base of grass clumps. It is inconceivable that non-residual moving clouds of aerosols will greatly affect these resting individuals.

Other population sampling techniques such as truck traps (Miura 1971, Nelson and Spadoni 1972) and light trapping or man-hour collection from resting sites were considered for pasture *Aedes*. Since the experimental areas treated were relatively small (160-500 acres), these sampling techniques were not employed. Landing counts of *Aedes* per human host was found to yield the needed information for the least amount of effort.

To assess the landing and biting activity of pasture *Aedes*, landing counts were taken by 3 or 4 individuals in treated and untreated fields in the same area at the same time. The counts were taken at several stations in each field. The sampler entered the field from one of the borders and walked 200-300 feet into the field and counted or estimated all mosquitoes landing on the lower extremities. Landing counts were taken prior to and at intervals after treatment.

Since the landing-biting activity of *Aedes* have diurnal rhythmicity (peak activity around sunrise and in the evening, declining activity after sunrise), it is very important that counts in untreated and treated fields are taken simultaneously to correct for any change in activity pattern due to natural causes. Once the data on the numbers of adults landing and biting were obtained, the means were calculated.

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The percent reduction was then calculated by Mulla's formula (Mulla et al. 1971) to compensate for the dynamism of populations in the treated and untreated areas.

Some uncontrollable factors which were observed to influence mosquito landing and biting counts were as follows:

1. Presence of cattle in the field drastically reduces landing-biting rates on the sampler. Cattle moving to a station with high level of mosquito activity caused a drastic decline in landing-biting activity. Such stations should be dropped from sampling.
2. Irrigation water reaching a given area of the field drives adult mosquitoes away from that portion of the field to another.
3. It is well known that damp and wet portions of fields have much higher density of adult mosquitoes than drier portions.
4. Ingress and egress of adult mosquitoes can cause substantial variability in mosquito density. This phenomenon, however, is not deemed to be operative over short periods such as sunrise to sunset.
5. Mowing or grazing of forage in a given area will eliminate resting sites for mosquitoes and thus result in lower density of adult mosquitoes.

**DROPLET SIZE DISTRIBUTION.**—One foot by three inch microscope glass slides were coated with magnesium oxide smoke and placed on the ground treated with propoxur OSC 1.4 formulation discharged at the rate of 6 oz/acre. In this experiment the slides were placed at right angle to the flight line of the plane in an irrigated pasture with ULV application of propoxur OSC 1.4 discharged at a pressure of 100 psi. In a second experiment a paraffinic oil (492 Process oil — Texaco) was sprayed at 10 oz/acre at

450 psi over slides placed on the ground in a bare field. Alternate slides were removed after discharging one swath over the slides and the remaining slides were removed after completion of second swath. Sufficient time was allowed between swaths to make sure slow-falling droplets reached the ground.

The diameter of the droplets was measured by a compound microscope supplied with a calibrated ocular micrometer. The frequency distribution of droplets by size groups in each test is plotted.

### Results and Discussion

**PYRETHRINS.** It is interesting to note that two similar applications of pyrethrin by air produced different results (Table 1). The first treatment where the theoretical swath was 660 feet and the delivered volume was 6 oz/acre, the level of adjusted reduction was higher than the second treatment where the theoretical swath width was reduced to 330 feet and the volume delivered was 12 oz/acre. This lack of efficacy could be due to lateness of the second treatment.

In the third treatment with pyrethrin, cotton seed oil was substituted for Klearol, which was not available. In this treatment the dosage of pyrethrin was increased to 0.02 lb/acre and the volume delivered was 8 ounces per acre. Adjusted reduction of *Aedes nigromaculis* (Ludlow) adults was very satisfactory in this treatment. It seems that both Klearol and cotton seed oil are satisfactory diluents for pyrethrin concentrates. The extent of reduction in applications using high pressure delivery system was slightly higher than that obtained with low pressure application equipment

Table 1.—Evaluation of pyrethrin (pyroicide intermediate 57; 10% containing 0.78 lb/gal of pyrethrin and 3.90 of piperonyl butoxide) applied by air against adults of the pasture mosquito *Aedes nigromaculis* (Rancho Santa Maria, Kern County, 1972).

Field	Acres	lbs/A	Fl oz/A	Carrier	Number Station	Mean No. landing/host (hours post-treatment)							
						Pre-treatment	1-2		2-3		3-4		
						No.	%R	No.	%R	No.	%R	No.	%R
5,6,7 <sup>a</sup>	320	.01	6	Klearol	4	70	11	80	4	90	—	—	—
4 <sup>b</sup>	160	.01	12	Klearol	6	132	120	0	116	0	72	0	—
7-12	320	Control	0	—	8	117	92	—	66	—	55	—	—
75 <sup>c</sup>	160	.02	8	Cottonseed Oil	9	183	14	91	12	90	—	—	—
76,74,69	480	Control	0	—	13	72	58	—	45	—	—	—	—

<sup>a</sup>This treatment was made on 6th of July between 0700 and 0800. The aircraft flew at 150 ft altitude and a swath width of 600 ft with a pressure of 200 psi.

<sup>b</sup>Treatment was the same as in <sup>a</sup> except the aircraft flew at 100 ft altitude and a swath width of 330 ft at 0830.

<sup>c</sup>This treatment was made on 15th of September starting at 0845 and ending at 0855. The wind was 2-3 mph from the WNW. The aircraft flew at 30-40 ft altitude and with a swath width of 330 ft, with a pressure of 450 psi on four nozzles. Pyrethrin conc. 6% and PBO 24%.

Table 2.—Evaluation of three experimental synthetic pyrethroids as applied by high pressure delivery system on an airplane to irrigated pastures against adult *Aedes nigromaculis*.

Field No.	Acres	Material	Fl. lbs/A	Carr- oz/A	ier	No. Sta.	Pre-treat	Mean number adults landing/host (hours post-treatment)							
								0-1		1-2		2-3		8-10	
								No.	%R	No.	%R	No.	%R	No.	%R
51	100	NIA 26021 <sup>a</sup>	.01	12	RLO & Light Med Oil	4	282	166	7	180	4	—	—	266	12
71	160	Control	0	0	—	4	256	162	—	170	—	—	—	275	—
51	80	NIA 18739 <sup>b</sup>	.05	12	Light Med Oil	6	7	9	0	4	0	10	0	18	0
71	160	Control	0	0	—	7	191	128	—	98	—	87	—	150	—
51	80	NIA 24110 <sup>c</sup>	.012	9	Process 492	6	775	76	90	46	94	33	95	134	76
53,71	30	Control	0	0	—	7	800	807	—	765	—	650	—	571	—

<sup>a</sup>NIA 26021 (10% solution containing 1 lb/gal of medium spray oil) was applied on 13th of September at 10:30 finishing at 10:40. The wind was 1-5 mph from north. Flying at 20-30 ft altitude and with a swath width of 200 ft, the aircraft had 5 nozzles open and a pressure of 450 psi.

<sup>b</sup>NIA 18739 (CRM-98, 10% solution containing 1 lb/gal of medium spray oil) was applied on 14th of September at 0915. The wind was 1-2 mph from the north. The aircraft flew at 20-30 ft altitude and with a swath width of 200 ft with 5 nozzles open and a pressure of 450 psi.

<sup>c</sup>NIA 24110 (10% solution containing 1 lb/gal of medium spray oil) was applied at 0800 finishing at 0810. The wind was 3-4 mph from the SW. The aircraft flew at 20-30 ft altitude and with a swath width of 200 ft with four nozzles open and a pressure of 450 psi.

(Mulla et al. 1971). In this analysis due consideration should be given to the higher rates of application evaluated in 1972 than in 1971 (Mulla et al. 1972).

**SYNTHETIC PYRETHROIDS.**—NIA 18739 or NRDC 107 or bioresmethrin-(5-benzyl-3-furyl) methyl (+)-trans-2,2-dimethyl-3-(2-methyl-1-propenyl) cyclopropane-1-carboxylate. This is trans component of NIA 17370 or resmethrin (SBP-1382).

NIA 26021 or NRDC 119 is the cis component of NIA 17370 or resmethrin or SBP-1382.

NIA 24110-(5-benzyl-3-furyl) methyl 1R,2R-2-[(cyclopentylidene)methyl]-3,3-dimethyl-cyclopropanecarboxylate.

These three synthetic pyrethroids related to SBP-1382 (resmethrin) were evaluated by applying the material in light medium paraffinic oils. These materials were furnished by Niagara Chemical Division, Food Machinery Corporation, Richmond, California. Since the quantity of the materials available was small, it was not possible to run numerous tests with each. Therefore, the information presented here is only preliminary in nature at this time, and further studies are needed on the efficacy of these synthetic pyrethroids.

NIA-18739 and NIA-26021 both did not produce any marked level of reduction (Table 2). The extent of coverage of the selected field was not adequate with these materials especially along the borders where counts are taken. Also the time of application being late in the morning could be responsible for the total lack of efficacy. The third analog, NIA-24110, proved to be highly effective. Since this material was used last, experience from the previous two materials precluded lack of good coverage, especially along the periph-

ery of the field where all sampling stations were established. Also the timing of the application which was earlier (8:00 a.m.) could have contributed to increased efficacy. Further studies are needed to assess fully the efficacy of these analogs against adult *A. nigromaculis* in irrigated pastures.

SBP-1382 or resmethrin or NRDC 104 or NIA 17370, a synthetic pyrethroid, containing 70% trans and 30% cis isomers of (5-benzyl-3-furyl) methyl-2,2-dimethyl-3-(2-methylpropenyl) cyclopropanecarboxylate, was evaluated against adult *A. nigromaculis*, using the high pressure delivery system. Two treatments were made at the rate of 0.02 lb/acre active ingredient and 2 other treatments at the rate of 0.01 lb/acre active material. The level of adult suppression was low to medium at the higher rates (Table 3). The lower rate, however, yielded much better control than the higher rate of application. These tests also showed better performance than those conducted in 1971 (Mulla et al. 1972). This is probably due to the timing of application. The two applications at 0.02 lb/acre were made rather late (about 9:00 a.m. or later). The two applications at the rate of 0.01 lb/acre were made much earlier (between 7-8 a.m.), a time when mosquito flying activity was probably high. According to Miura (1971), *A. nigromaculis* has bimodal flying activity with peaks occurring near sunrise and at dusk. Other factors such as inversion and turbulence could also influence adulticidal efficacy in irrigated pastures.

SBP-1382 performed much better when it was applied by a non-thermal aerosol to irrigated pastures (Sjogren et al. 1973) and presented in an accompanying paper in this issue.

**PROPOXUR, PRIMIPHOS METHYL AND CIDIAL.**—Ultra low volume application of a new formulation of pro-

Table 3.—Evaluation of SBP-1382 (25% concentrate) applied by high pressure system on an aircraft against adults of the pasture mosquito *Aedes nigromaculis*.

Field No.	Acres	Dosage		Carrier	No. Sta.	Pre-treat	Mean number landing/host (hours post-treatment)							
		lb/ac	Fl. oz/A				0-1		1-2		2-3		8-10	
							No.	%R	No.	%R	No.	%R	No.	%R
53	160	0.02 <sup>a</sup>	8	ARCO LO	6	230	112	21	120	20	212	12	—	—
71	160	Control	0	—	4	262	162	—	170	—	275	—	—	—
53	160	0.02 <sup>b</sup>	12	Cottonseed	4	144	—	—	30	78	43	58	176	0
71	160	Control	0	Oil	7	140	—	—	130	—	100	—	150	—
16-17	480	0.01 <sup>c</sup>	8	ARCO LO	7	16	1	83	1	79	1	75	—	—
20	160	Control	0	—	4	24	9	—	7	—	6	—	—	—
4-10	450	0.01 <sup>d</sup>	8	ARCO LO	8	298	23	92	20	90	—	—	22	85
53 & 71	320	Control	0	—	4	84	80	—	56	—	—	—	41	—

<sup>a</sup>Treatment was made Sept. 13 at 0958. The aircraft flew at 20-30 ft altitude with a swath width of 320 ft. Five nozzles were used with a pressure of 450 psi.

<sup>b</sup>Treatment was made Sept. 14 at 0850. The aircraft flew at 20-30 ft with a swath width of 200 ft. Six nozzles were used with a pressure of 450 psi.

<sup>c</sup>Treatment was made August 22 between 0700 and 0755. The aircraft flew at 40-50 ft altitude and a swath width of 320 ft. Five nozzles with a pressure of 450 psi.

<sup>d</sup>Treatment was made August 23 between 0705 and 0755. Six nozzles open, pressure 450 psi.

Table 4.—Evaluation of various adulticides in irrigated pastures against *Aedes nigromaculis* by ULV applications using high pressure delivery system on an aircraft (Rancho Santa Maria, Kern, Tulare, Kings Counties, California, 1972).

Field No.	Acres	Material	lbs/A	Fluid oz/A	No. Sta.	Pre-treat	Mean number landing/host (hours post-treatment)							
							0-1		1-2		2-3		24	
							No.	%R	No.	%R	No.	%R	No.	%R
75	160	Baygon <sup>a</sup>	.06	6	6	141	60	56	36	69	16	84	—	—
53,71	320	Control	0	0	7	800	765	—	650	—	550	—	—	—
12	160	Actellic <sup>b</sup>	.025	6	6	145	65	49	76	49	59	43	117	0
4,5,6	160	Control	0	0	6	105	92	—	107	—	75	—	70	—
City Sewer Farm	160	Cidial <sup>c</sup>	0.125	8	8	38	—	—	1	97	—	—	—	—

<sup>a</sup>Baygon OSC (1.4), 7.5 gallons placed in aircraft. Treated September 27 at 0900 finishing at 0915. The wind was 3-5 mph from the west. Four nozzles were used flying at 30-40 ft altitude with 100 psi pressure.

<sup>b</sup>Actellic (ULV 5 lb/gal) was applied on July 5 at 0815 hours finishing at 0821. Nine nozzles were used flying at an altitude of 100 ft and a swath width of 660 ft with a pressure of 100-300 psi.

<sup>c</sup>Cidial EC<sub>4</sub> diluted 1:1 with paraffinic larvicidal oil. Pressure 450 lb/in<sup>2</sup>. Application made on September 18 at 0900.

poxur with the high pressure delivery system (although the pressure maintained was lower than expected) yielded a medium level of reduction in adult activity (Table 4). This level of performance is inferior to the application of this material by a ground non-thermal aerosol generator (Sjogren et al. 1973).

The droplet spectrum obtained with the aircraft system discharging propoxur OSC 1.4 was quite coarse (Fig. 1), bulk of the droplets were in the range 30-100 microns. In the second study of droplet size spectrum using a paraffinic oil (492 Process Oil), the spray was delivered at higher pressure (450 psi) and as can be seen the droplet size was con-

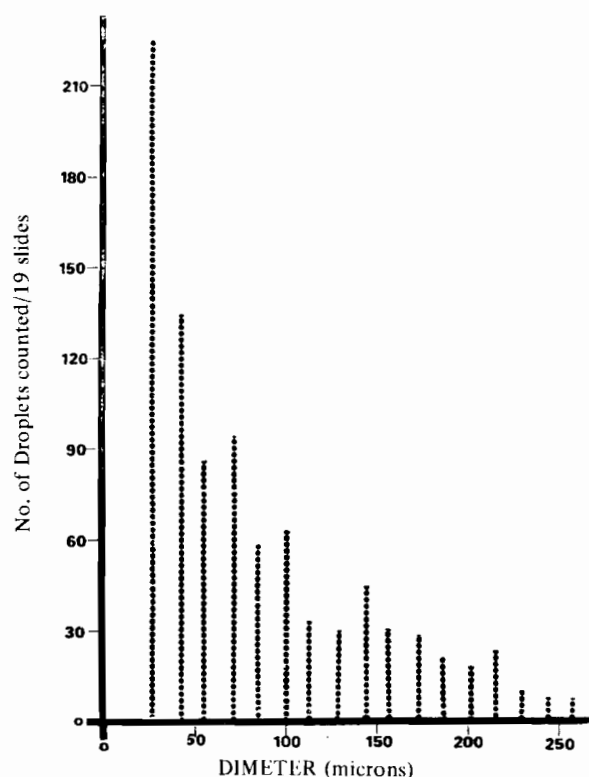


Fig. 1.—Frequency distribution of droplets of propoxur OSC 1.4 formulation discharged at the rate of 6 oz/acre from a high pressure delivery system on an aircraft. Pressure during application was 100 psi.

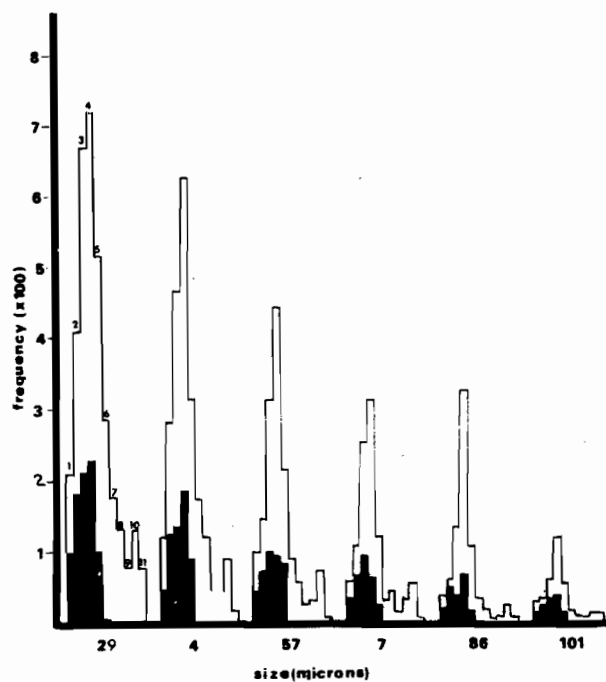


Fig. 2.—Frequency distribution of droplets of 492 process oil discharged at the rate of 10 oz/acre from a high pressure delivery system on an aircraft when deposits were made from 1 and 2 swaths. Shaded areas 1 swath, open areas total of 2 swaths. Slides 1-10 from north to south.

siderably smaller (Fig. 2). The number of droplets deposited increased considerably when the second swath droplets were also deposited on the slides exposed to the first slide. This disproportional increase was due to displacement of the first swath northward due to breezy conditions during the application of the first swath.

In the propoxur treatment the average number of all sizes of droplets was less than  $0.25/\text{cm}^2$ . In the treatment with 492 Process Oil the average number of droplets for positive slides (those having deposit) was about  $36/\text{cm}^2$  for the one swath and  $64/\text{cm}^2$  for two swaths. There were more slides showing deposits after the second swath than after the first swath.

Primiphos methyl or Actellic, (2-diethylamino-6-methyl-pyrimidin-4yl-dimethyl phosphorothionate), at the rate used produced a mediocre level of reduction of adult mosquitoes. Cidal or phenthoate, (0,0-dimethyl dithiophosphoryl-1-phenylacetate), at relatively high rate yielded good reduction in adult activity.

All three applications in these series were made late in the morning. It is possible that proper timing of application either early in the morning or at dusk will produce better results than those obtained here.

CONCLUSIONS.—Some tentative conclusions can be drawn from these preliminary studies.

1. Both pyrethrin and SBP-1382 could suppress adult mosquitoes in irrigated pastures at the rate of 0.01 lb/acre applied from air.
2. Other synthetic pyrethroids also hold promise and could yield good mosquito control at 0.01 lb/acre applied from air.
3. Some organophosphorus insecticides could suppress adult *Aedes* mosquitoes when applied at 0.05-0.1 lb/acre by air.
4. There are indications that aerial treatments should coincide with peak flight activities of mosquitoes which occur at sunrise and early evening hours.
5. Aircraft altitude during ULV applications should be held to under 50 feet above ground and swath widths of 300 feet or so should be established.
6. The high pressure delivery system appeared to produce smaller droplets than the low pressure system. Further work is necessary to establish this fact.
7. Thorough treatment of field borders and periphery is essential for good results. The field should be bordered with swaths according to the wind direction. Since most population assessment counts are taken along the borders of a field, nontreatment of field periphery could yield erroneous information.

#### Acknowledgment

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## INSECT DEVELOPMENT INHIBITORS: THEIR POTENTIAL AS MOSQUITO CONTROL AGENTS

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### ABSTRACT

A slow-release formulation of ZR-515 (isopropyl 11-methoxy-3, 7, 11-trimethyl-2, 4-dodecadienoate) has proven to be practically feasible for controlling field populations of OP-resistant strains of *Aedes nigromaculis*: successful results on pastures were obtained with aircraft applications using doses as low as 0.025 lb AI/acre. Aircraft applications of 0.1 and 0.05 lb AI/acre of the same formulation also

gave encouraging results against populations of *Aedes melanimon* larvae on duck club ponds. Results on populations of *Culex tarsalis* were not considered operationally feasible and further research need is indicated. Laboratory and field evaluations of Ro20-3600 (6, 7-epoxy-3-methyl-7-ethyl-1-[3-4-(methylenedioxy) phenoxy]-2-cis-trans-octene) and R-20458 (trans-8-(4-ethylphenoxy)-2, 6-dimethyl-2-3-epoxy-6-octene) showed lesser promise against *A. nigromaculis*.

# A COMPARISON OF CONCENTRATED INSECTICIDE SOLUTIONS AND EMULSIONS FOR ULTRA LOW VOLUME APPLICATION AGAINST MOSQUITOES AND HOUSE FLIES

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Field tests were conducted during August and September 1972 to determine the efficacy of concentrated insecticide emulsion aerosols for the control of adult mosquitoes and house flies. The applications were made with an aerosol generator developed at the Navy Disease Vector Ecology and Control Center, Alameda. Twenty-eight tests were conducted with the flow rate for the majority in the range of 29 to 33 ounces per minute and a vehicle speed of five miles per hour. The insecticides used in the study included fen-thion 1 lb/gal, chlorpyrifos  $\frac{1}{4}$  lb/gal and naled 3 lb/gal. The selection of a particular insecticide concentration was somewhat arbitrary since the purpose of the study was to compare emulsions and solutions with no attempt to compare insecticides. Cottonseed oil was used as the diluent for the oil solution concentrates.

Adult mosquitoes (*Culex pipiens*) and flies (*Musca domestica*) were exposed in cages three feet above the ground and along a line perpendicular to the path of the aerosol generator. The cages were placed at intervals of 500 feet and the line extended to 5000 feet. Control stations were located approximately one mile upwind. Mortality was determined 24 hours after exposure. The study was conducted at Skaggs Island, a naval reservation located at the northern tip of San Pablo Bay. All tests were conducted during the day between 1030 and 1500 hours. Meteorological condi-

tions at Skaggs Island were near ideal. The wind direction was very consistent and the velocity usually ranged from 5 to 12 knots. A temperature inversion usually existed at the site during the test periods.

Results of the tests consistently demonstrated that emulsions were about equal to oil solutions. This was observed with all three insecticides. Swath widths to 5000 feet with 100 percent mortality in the mosquitoes were easily achieved during the tests. Considering that the water would evaporate within a few seconds following generation of the droplet it is quite likely that a significant percentage of the droplets were reduced in size to 3 to 5 microns before reaching the target. This particularly would apply to fen-thion where approximately 90 percent of the total volume was water.

It is conceivable that the smaller droplets would be adversely affected by lapse temperature conditions. However, this should not present a real problem since most adult mosquito control operations are conducted at times when an inversion exists. The lower costs, reduced environmental contamination by diluents, ease of cleaning equipment and general convenience constitute advantages that make emulsions worth considering for ULV ground applications.

## AEROSOL MACHINE DEVELOPMENTS

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Increasing concern over the reduced effectiveness of mosquito larviciding, primarily because of larvae resistance to many chemicals, has prompted the MAD managers in California to look more and more toward adulticiding with aerosol drop size sprays for rapid knock-down and thereby primary protection of urban population centers.

Aerosoling may be defined as applying sprays with VMD (volume median diameter) of less than 50  $\mu\text{m}$  by aircraft, or under 25  $\mu\text{m}$  VMD by ground equipment. Since these sprays are by definition airborne the technique of aerosoling requires much greater care and precision in the application machine, methodology, and the correct weather or micro-meteorological conditions in order to achieve predictable success. If care in the application process is not pursued, the results may be erratic, unpredictable and frequently wasteful of time, material and labor.

**AEROSOL DROP SIZE.**—Probably the most important parameter or variable function that must be controlled when aerosoling is the drop size of the aerosol. The atomization process is one of tremendously expanding the surface area of a given quantity of liquid. Thus, producing 100  $\mu\text{m}$  drops (or an atomization process that produces a VMD of 100  $\mu\text{m}$ ) demands a given level of energy input as liquid pressure, or air volume under pressure or rotary motion, heat or other source of energy. Reducing the drops to a VMD of 50  $\mu\text{m}$  does not just double the energy requirement, but increases it by 4 times because the new surface area created is ( $d^2$ ) or 4 times that of the 100  $\mu\text{m}$  drops.

But this is only part of the story. In addition to the energy again, but because the smaller drops are also more resistant to break up (that is small drops require greater energy to split than larger ones) we have quickly reached a point where the normal air-shear or force of liquid ejected at high pressure from a nozzle will no longer break up the drops and other more powerful means must be used. Thus, we can use such means as the following for obtaining a breakup of spray liquids (either oil or water base) down to around 60-75  $\mu\text{m}$  VMD. But to go below this requires different procedures as shown:

Atomization means to reduce spray liquids to approximately minimum of 60  $\mu\text{m}$  VMD.

1. Hydraulic spray pressure and hydraulic fan or cone type nozzles. Pressure above about 250-300 psi (pounds per sq in).
2. Rotary devices, Micronaire, Becomist, Minispin, or others with a disc or screen peripheral speed approximately 15,000 fpm (ft. per min.).
3. Air-shear or twin fluid systems using 200 to 300 mph air velocity or relative velocity between emitted spray and surrounding airstream, such as with a blower sprayer or an aircraft.

Systems required for producing aerosols or sprays under 50  $\mu\text{m}$  VMD.

1. High pressure air/liquid or 2 fluid nozzles such as external or paint sprayer type device or internal as high pressure nebulizer nozzles.
2. High air volume at low pressure systems such as the Yeomans-Belvoir system (also Leco, Microgen, etc.) using 100-200 cfm (cubic ft/min) of air at 3-5 psi in a vertical type nozzle.
3. Liquid flashing or pressurized aerosol (as Freon pressurized cans) where a highly evaporative carrier is mixed under pressure with the spray liquid and when released forms aerosol size drops, further reduced by the evaporation or flashing (changing from liquid to gaseous state) of the spray toxicant.
4. Thermal or heat generated aerosols where atomization is produced by the "flashing" of the liquid toxicant by direct contact with a heated surface or by mixing with steam. Aerosol size toxicant particles will tend to cool in the moist atmosphere and produce considerable amounts of undesirable dense fog or smog associated with the thermal machines.

**METEOROLOGICAL CONDITIONS.**—Where the aerosol size particles (below 50  $\mu\text{m}$  VMD) are to be used, great care must be taken to apply these only under favorable weather conditions conducive to proper surface downwind spreading with minimum vertical or turbulent mixing and loss to upper air. It is extremely difficult to predict the success of aerosoling by aircraft if drops less than 50-60  $\mu\text{m}$  VMD are used. This is because of the height of release above brush or forest canopy which subjects the aerosol to overhead dispersion in the air from the aerodynamic Vortex system of the aircraft.

Thus, aerosols below 50  $\mu\text{m}$  VMD, which are most successful in contacting adult mosquitoes, are not easily handled by aircraft and so applied are subject to erratic biological results. Drops down to 10  $\mu\text{m}$  VMD have been shown to be very successful in contacting flying mosquitoes. These can be produced by a variety of ground operated machines, and many observers have had very good results applying these aerosols with drop size containing about one minimum  $\text{LD}_{50}$  dose for a mosquito.

For aircraft adulticiding it is suggested that the VMD be not smaller than 50  $\mu\text{m}$ . Even with this coarse aerosol applied under best weather conditions the success of the operation can frequently be spotty as the job is completely dependent on favorable weather conditions. Aerosoling offers a very effective knock-down tool for district managers and new cold fog aerosol type equipment not only eliminates the undesirable smoke of the thermal machines but also reduces the losses of the material that went into the inefficient sub-micron size smoke particles. Programs for adulticiding large urban areas have shown the protective potential of these techniques, but where heavily infested and favorable breeding areas exist the adulticiding program will show very little residual effect and must be repeated for continued control.

# OPERATIONAL EXPERIENCE WITH LOW VOLUME NONTHERMAL AEROSOL GENERATORS IN THE COLUSA MOSQUITO ABATEMENT DISTRICT

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**INTRODUCTION.**—The Colusa Mosquito Abatement District includes an area of 102,400 acres in the Sacramento Valley of California, much of it in agriculture. The major population center is the City of Colusa, population 3,825. Mosquito problems include *Aedes* species from wildfowl management areas, private duck clubs, and irrigated pastures, and *Anopheles freeborni* and *Culex tarsalis*, produced in rice fields. Within the District, about 20,000 acres are usually planted to rice. There are an additional 70,000 acres of rice in the adjacent uncontrolled area of Colusa County as well as rice growing areas in neighboring counties which are not covered by mosquito control programs.

Since the formation of the District in 1958, most of the control effort has been directed against *Aedes* species. This has provided the citizens with protection during the daylight hours, but *A. freeborni* and *C. tarsalis* from the rice fields have continued to cause extreme discomfort and constitute a continuing threat of disease transmission. Over the years, attempts were made to utilize parathion to control mosquitoes in rice, based on inspection and treatment of individual fields as deemed necessary. The technique proved to be unsuccessful because of the magnitude of the rice acreage and the difficulty in sampling for larvae in that habitat. Light trap counts showed little difference between years when treatment was attempted and years when it was not. In 1967, a large scale experimental application was made as a cooperative study to evaluate the potential of aerial larviciding with low volume application of chlorpyrifos (Dursban®). In 1970, the technique was incorporated into the District's operational program. The work was described by Womeldorf and Whitesell (1972), Akesson et al. (1972) Washino, Whitesell et al. (1972), Washino, Ahmed et al. (1972) and Atkins (1972). The conclusion reached was that, although the technique killed larvae, the degree of control afforded was not satisfactory because of reinfestation from the vast area of uncontrolled rice fields surrounding the District.

**DESCRIPTION OF MACHINES.**—Tests with low volume nonthermal aerosol generators reported by Stains et al. (1969) and the operational experiences of neighboring mosquito abatement districts encouraged construction and trial of similar units in the Colusa MAD. A thermal fog unit was converted to a coldfogger similar to the conversion described by Anderson and Schulte (1970). The unit consisted of a two-cylinder, 12 hp Wisconsin engine belt-coupled to a positive displacement air pump (Miehle-Goss-Dexter). Air from the blower was ducted to a nozzle manifold in which were mounted four Afa No. 51140 venturi airshear nozzles. The insecticide container was pressurized with air bled from the blower. With this unit we were able to obtain a pressure of approximately 3 psi in the nozzle manifold and a maximum insecticide output of 5 fl oz/min. Although droplet size measurements were not made with this configuration, gross

observations led us to believe that the air output was insufficient to provide adequate atomization with four nozzles. Two of the nozzles were removed which resulted in increased pressure in both the nozzle manifold and the insecticide tank. Output remained about the same with an apparent decrease in droplet size. Droplet size measurements made by Metronics Associates Inc., Palo Alto, California, using a fluorescent particle technique, showed that most droplets were in the 6-10  $\mu$  range (L. M. Vaughn, personal communication). This unit was used experimentally in operational applications during the 1970 and 1971 seasons.

During the winter of 1971 a new machine was constructed (Figure 1). It consisted of a 21 hp Crosley engine direct-coupled to a Roots No. 47 blower. The nozzle used was an Afa No. 6208-208A triple-venturi assembly which had been modified by inserting brass tubing into the orifices to reduce the size of the opening to 1/32 inch. The nozzle manifold was constructed with a removable face plate. Face plates containing 1 to 4 nozzle assemblies were tested. It was decided that the plate containing two of the triple-venturi assemblies was the most satisfactory, apparently providing a good balance between number of nozzles and the amount of air produced by the blower at moderate engine speeds. No precise droplet sizing was done at the time, but visual observations were made of the spray and droplet fallout checks were made with the aid of aluminum pie plates. Measurements carried out later by the Department of Agricultural Engineering, University of California at Davis, indicated that the machine was producing droplets in the range of 16  $\mu$  VMD and 24.5  $\mu$  VMD (N. B. Akesson, personal communication).



Fig. 1.—Colusa Mosquito Abatement District nonthermal aerosol generator. 1971 model.

**FIELD TRIAL RESULTS.**—Experimental operational applications were made in four types of areas: the town itself, the rice fields around the populated area, pastures, and duck clubs. Experimental applications were made in the town of Colusa during the 1972 mosquito season using 3% pyrethrin synergized with 25% piperonyl butoxide, and 1 lb/gal chlorpyrifos. Details of these tests are given by Womeldorf et al. (1973). Mortalities of caged adult mosquitoes were much greater on test-line applications run outside of town in open terrain than observed mortalities in town with the same chemical formulation and discharge rate.

Several experimental applications were made to rice areas with the new direct-coupled units. Chlorpyrifos was used at rates of 1 lb/gal active ingredient and 2 lb/gal active ingredient. These formulations were prepared either by combining Dursban ULV with an equal amount of polypropylene glycol (Dow P400®) or by diluting the 6 lb/gal thermal fog formulation with a suitable diluent such as Sontex 75-T, Niagara Orchem or Occidental Chemical Co. Super 94.

Swath widths of 1, 2, and 3 miles were attempted. Test areas of from 6 to 8 square miles were treated at each dosage rate. No noticeable reduction in adult resting unit counts was obtained in the test areas so we decided to attempt treatment of an area large enough to prevent rapid reinfestation. Two identical units were used on the test which covered approximately 70 square miles. Operational time was about 4 hours during which 240 lbs of chlorpyrifos were dispersed. Results were unsatisfactory, possibly due to assuming swaths too wide under unfavorable wind conditions.

A large duck club area of 12,000 acres on the eastern side of the District is initially flooded during early August through mid-October. Most of the *Aedes* species production is controlled by low volume aerial larviciding, but complete coverage is not possible. Nonthermal aerosols have been very effective in knocking down emergent populations in relatively small areas.

**CONCLUSIONS.** Our experience with coldfoggers over the past two seasons has indicated that they are a useful tool under particular situations. The machines have been very valuable in the District's *Aedes* species control program, especially in the duck club areas.

Aerosols alone have not enabled us to achieve an acceptable level of control over *A. freeborni* and *C. tarsalis* from rice fields, nor has wide-swath, low-volume larviciding proved a complete success. The area involved is so large and so

inaccessible that thorough coverage is difficult. Mosquito production in the uncontrolled rice surrounding the District is of such magnitude that control efforts are overwhelmed by the influx of "foreign" mosquitoes. It is possible that a combination of the two techniques, coupled with the use of available naturalistic control measures, might be effective if sufficient area could be covered. At present this integrated approach is beyond the financial capabilities of the District.

The macro- and micro-climatological conditions of the in-town environment are sufficiently different from those of open country to necessitate changing some basic operational approaches. Further tests are needed before we can determine if the technique will be successful in controlling mosquitoes in urban areas.

Our future work with low volume nonthermal aerosols will emphasize the relationship of swath widths to insecticide concentrations and discharge rates, the effects of various weather conditions, and a continued search for the most effective insecticide.

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# EVALUATION OF MOSQUITO ADULTICIDES APPLIED AS NONTHERMAL AEROSOLS IN IRRIGATED PASTURES

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**INTRODUCTION.**—The increased reliance upon adulticides by mosquito abatement agencies encountering control failures with organophosphate larvicides has directed greater attention to evaluation of adulticidal compounds (Mulla et al. 1972) and techniques of their application. Districts limited by OP resistance in mosquitoes to most registered larvicides during 1971-72 seasons, found only propoxur (Baygon®) and dichlorvos (DDVP) effective adulticides, the latter only under certain meteorological conditions (Ramke et al. 1969, Sjogren 1973).

Control of susceptible adult mosquito populations has been successful in past years with ULV systems. With such systems a large percent of the spray volume is contained in the >150 VMD droplet range, which unless encountered after deposit by adults moving within the field, is wasted. In the case of non-persistent adulticides such as propoxur, SBP-1382, pyrethrins and others, it is necessary that a great portion of the adult mosquitoes be hit by the insecticide delivered as an aerosol cloud traveling over the infested area. Fortunately, the technology necessary to produce the required droplet range has been developed in recent years in the form of the low-volume aerosol generator (Stains et al. 1969) and recently a high pressure delivery system for aircraft was designed and tested under field conditions (Mulla et al. 1973).

This paper will report the results obtained with four adulticidal compounds applied with a cold fog generator against OP resistant *Aedes nigromaculis* on the Rancho Santa Maria (Smiths) pasture complex in northwest Kern County, California, during 1972.

**MATERIALS AND METHODS.**—Pyroicide 57 (10% pyrethrins plus 50% piperonyl butoxide) concentrate was applied straight or diluted with paraffinic petroleum hydrocarbons such as Klearol (Witco Chemical Co.) or 492 Process Oil (Texaco Inc.) SBP-1382 (NRDC 104 or NIA 17370) [(5-benzyl -3- furyl) methyl 2,2-dimethyl -3- (2=methyl propenyl) cyclopropanecarboxylate - (approx, 70% trans; 30% cis isomers)] 2 lb concentrate was applied in raw cottonseed oil and ARCO Larvicide Oil (Atlantic Richfield Co.), an aromatic oil. Propoxur (Baygon) (O-Isopropoxyphenyl methylcarbamate) 1.4 oil soluble concentrate was applied undiluted. Primiphos methyl or Actellic® (2-diethylamino - 6 - methylpyrimidin - 4 - yl dimethyl phosphorothionate) 5 lb ULV and EC (emulsifiable concentrate) concentrate was mixed with 492 Process Oil. The three petroleum hydrocarbon carriers are characterized with high boiling ranges.

All applications were made with a four nozzle Microgen® (Model L 4-75) cold fogger transported on the tail-gate of a pickup truck. Route of travel was wind determined with the truck traveling on the upwind side as near right angle to the field as possible. Rate of travel averaged 5 mph.

Problems were encountered with the use of the Microgen model employed in these tests. The lack of a drain valve on the insecticide reservoir made it necessary to empty residual fluid out the nozzle after each test. The machine weight and method of tank attachment prevented tipping. Similarly, it was not possible to determine the tank volume during a run, without stopping, depressurizing and dipping with a stick. An external fluid column would enable better use of the last few gallons by making possible alterations in the final minutes of treatments. Since engine RPM alters the pressure delivered by the blower, a locking throttle mechanism would prevent flow rate changes during operation caused by vibration or accidental movement. An air pressure gauge inserted between the blower and the nozzle chamber via a quick-disconnect coupling attachment would permit engine throttle adjustment to achieve a constant air pressure (K. G. Whitesell, Personal Communication).

Wind direction changes during treatment made proper coverage of the test area difficult or uncertain, frequently requiring tests to be qualified or voided. Wind direction during twilight periods could not be predicted for any period of time in areas surrounded by heterogeneous ground covers, whereas, experience showed air flow direction stable for up to an hour after sundown approximately 70% of the time when the treated area was encompassed for several miles by homogeneous ground. Contrary to results obtained in the Sacramento Valley (K. G. Whitesell, Personal Communication) at wind speeds up to 6 mph, treatments in Kern County at air velocities above 4 mph produced little or no decline in *Aedes* populations. Suppression of flight activity by winds above 4 mph, or a requirement of multiple particle contact by OP resistant adults to produce mortality, or both, may have influenced results.

Fields chosen for treatment were flood irrigated quarter and half mile Bermuda grass pastures with a history of heavy *Aedes* production and resistance to OP larvicides. Test (pre & post evaluation) area size ranged from 320 to 640 acres. Adult populations were estimated from lower body landing counts taken pre and post treatment from locations within both the treated and control areas. The effect of of adult trailing behavior was minimized by traveling within a closed vehicle before re-entering the field for each subsequent count. Pre-treatment counts were usually made 5-45 minutes before treatment, post-treatment population assessments varied depending on time of treatment (i.e. lighting conditions) and material applied. Declines or increases in the adult population during each post-treatment evaluation period were corrected by Mulla's formula (Mulla and Darwazeh 1971) to more accurately determine the degree of

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suppression due to the adulticidal chemical.

The daily flight rhythms of *A. nigromaculis* in the San Joaquin Valley have been reported by Miura (1971) who found the maximum adult population in flight during the evening twilight period when the temperature is above 60°F and the relative humidity is 40% or higher. Experience has shown that cold fog particles drift over open ground without contacting adult *Aedes* resting in low growing pasture grasses. Thus applications were primarily made during periods of morning or evening flight activity. Wind velocity during the applications ranged from 0-6 mph.

**RESULTS AND DISCUSSION.**—On the evening of July 11, a cooperative test was conducted with Commander Ed Fussell, USN, Alameda, using a Wankel driven Microgen cold fogger to discharge propoxur (1.5 ec) at a flow rate of 55 fluid ounces/minute. Due to an abbreviated run, post-treatment counts were made in the estimated center of the swath, accounting for wind deflection. Essentially 100% reduction was observed in the adult population at distances up to a mile (Table 1). Recalling an adult count taken for operational purposes the morning of treatment approximately 3½ miles in line of drift downwind, a post-treatment count indicated substantial reduction. During this trial, control fields were not established for the assessment of population activity.

Table 1.—Initial application of propoxur (1.5 EC) applied by Microgen cold fogger against adult *Aedes nigromaculis* at Rancho Santa Maria (7/11/72, evening)<sup>a</sup>.

Field	Dist. from Discharge	Landing Rates / Host		% control <sup>b</sup>
		Pre(7:45pm)	Post (12 hrs)	
75	0.5 mile	1,000	0	100
69	1.0 mile	1,500	1	100
68	1.0 mile	150	0	100
68	1.3 mile	150	1	99
44	3.5 miles	60	11	82

<sup>a</sup>Three gallons of undiluted propoxur (1.5 EC) was dispensed at 55 oz/min 0.5 mile run from 8:22 - 8:29. Wind consistent from N. W. at 3-4 mph. Temperature 78 F.

<sup>b</sup>Comparisons not made with control fields.

On July 18, following cancellation of a planned high pressure aircraft adulticiding system test due to mechanical difficulties, a mid morning cold fog application was made to fields 4 through 10 on which pre-treatment counts had been taken. Ten gallons of pyrethrum 0.9% was applied in Klearol carrier at a flow rate of 24 fluid ounces/minute over approximately 500 acres. Wind velocity was 2-4 mph. Within the first 100 feet downwind 86% reduction was observed two hours post treatment, with control falling off to 23% at 200 feet, 43% at 500 feet, 52% at 1320 feet and 40% at just under 3960 feet, for an overall 49% reduction.

The following morning adult counts were again high in fields 4-10 and 5 gallons of 5% pyrethrum were discharged at a flow rate of 25 ounces/minute under a 4-6 mph breeze starting at 7:15 a.m. The control level increased slightly from the 1 hour post-treatment assessment to that taken 2

hours after treatment, 86% to 96% at distances up to 500 feet. The level of control dropped to 12% at 2,500 feet from discharge.

On the evening of August 18, a treatment of 5 gallons of 5% pyrethrum was made with a 1-3 mph breeze starting 10 minutes after sundown. For unknown reasons, the post-treatment results were variable, more so within the treated area than the control.

On the evening of September 14, an undiluted application of 5 gallons of 10% pyrethrum was made on a 1.25 mile run at a flow rate of 43 fluid ounces/minute, beginning shortly after sunset. Wind velocity was 1-2 mph. Twelve hour post-treatment counts indicated good population reduction with 98% control at 200 feet, 70% at 1,300 feet and 83% at 2,500 feet downwind (Table 2).

In these tests the poor control results achieved beyond 100 feet distance downwind from discharge in the 0.9% and 5% morning pyrethrum applications are probably influenced by insufficient flight activity. As Miura (1971) demonstrated, the evening adult flight activity is approximately twice as great as that in the early morning. Since cold fog particles do not penetrate thoroughly into grasses and other ground level adult resting areas, adult flight is essential for contact with insecticide particles. The inconsistent results gained from the 5% evening treatment may have been due to post-treatment infiltration of adults into the treated area, wind shift, or a combination of inadequate dosage and vigor tolerance conferred by acquired OP resistance. When the fogging solution was raised to 10% pyrethrum and flow rate increased from 25 to 43 fluid ounces/minute relatively good control was observed up to 2,500 feet downwind.

The first of three evening cold fog applications of SBP-1382 was made on August 22, across a 640 acre test area. Twelve and a half gallons of 4.8 SBP-1382 was applied at 106 ounces/minute in ARCO Larvicide Oil in a 2-3 mph breeze. Control assessment made 12 hours posttreatment reflected a mediocre level of control at 0.5 and 1 mile of 65% and 69%, respectively.

The second application was made in mid-September with 10 gallons of 7.2% SBP-1382, using raw cottonseed oil as carrier. The treatment commenced approximately a half hour after sundown at a flow rate of 28 fluid ounces/minute. Under changing wind conditions which necessitated treating the 500 acre plot on three sides, fair to good control was obtained at all stations. The distance from discharge to each station is estimated due to uncertainty as to which side or sides treated produced the population reduction. Control level varied from 97% to 53% within the evaluation area.

The following evening a 320 acre area was treated with 4 gallons of 7.2% SBP-1382 at 30 fluid ounces/minute flow rate. Approximately 12 hours post treatment, the control level averaged 71% at 200 feet, 53% at 1,050 feet, 61% at 2,500 feet and 45% at 4,200 feet (Table 3). Each of the SBP-1382 treatments was made under excellent conditions, with temperatures above 70°F, low wind conditions, and high mosquito flight activity.

Nozzle plugging was encountered in preliminary tests with propoxur (1.5 ec) applied diluted with ARCO LO,

Table 2.—Effectiveness of pyrethrins (pyroicide intermediate 57; 10%, containing 0.78 lbs/gal of pyrethrin and 3.90 lb/gal of piperonyl butoxide) diluted with paraffinic oils and applied by a cold fogger against adults of the pasture mosquito *Aedes nigromaculis* (Rancho Santa Maria, 1972)<sup>a</sup>.

Field	Acres	Date	Time	Vol. Fluid oz/min	% Pyre- thrins	Car- rier	Dist. from disch. (ft)	Pre- trt.	Mean number landing per host (hours post treatment) <sup>b</sup>									
									0-1		1-2		2-3		10-12		12-15	
									No.	%R	No.	%R	No.	%R	No.	%R	No.	%R
4-10	500	7/18	0930 1040	25	.9	Klearol	100	225	45	87	35	86	-	-	200	11	-	-
							200	148	151	32	142	23	-	-	34	77	-	-
							500	82	73	41	58	43	-	-	50	39	-	-
							2,500	127	120	37	76	52	-	-	260	0	-	-
							3,700	100	75	50	75	40	-	-	-	-	-	-
51	80	7/18	--	control	0	--	100	150	-	125	-	-	100	-	-	-	-	
4-10	500	7/19	0715 0750	25	5	Klearol + 492	100	515	80	82	25	93	25	93	-	-	-	-
							500	10	2	77	0	100	2	72	-	-	-	-
							2,500	112	125	0	70	12	70	12	-	-	-	-
51	80	7/19	--	control	0	--	87	75	-	62	-	62	-	-	-	-		
4-10	500	7/18	2000 2020	25	5	492	100	287	-	-	-	-	-	515	0	-	-	
							500	50	70	0	30	46	-	-	10	82	-	-
51	80	7/18	--	control	0	--	100	125	-	112	-	-	113	-	-	-		
53,71	320	9/14	1940 1955	43	10	--	200	221	-	-	-	-	-	5	98	1	99	
							1,200	75	-	-	-	-	-	20	70	35	27	
							2,500	96	-	-	-	-	-	15	83	16	74	
4,7,8	320	9/14	--	control	0	--	118	-	-	-	-	-	106	-	76	-		

<sup>a</sup>2-mile runs made in each test except on September 14 when the run was 1.25 miles.

<sup>b</sup>Counts were taken at 2-8 stations at each interval.

Table 3.—Effectiveness of SBP-1382 (conc. solution 2 lbs/gal diluted with carrier) applied by cold fogger against adults of the pasture mosquito *Aedes nigromaculis* (Rancho Santa Maria, 1972).

Field	Acres	Date	Time	Run miles	Rate fl oz/min	% SBP 1382	Carrier	No. sta.	Dist. from disch.(ft)	Pre- treat	Mean number landing per host			
											0-1		10-12	
											No.	%R <sup>a</sup>	No.	%R
68,69 75,76	640	8/22	2000 2030	2.5	106	4.8	ARCO-LO	3	2,500	77	-	-	14	65
								4	5,200	113	-	-	18	69
74	80	8/22	--	-	control	0	--	4	--	48	-	-	25	-
4 to 10	500	9/12	1935 2001	2.5	28	7.2	cottonseed oil	3	200	210	-	-	15	92
								2	1,050	108	-	-	20	79
								2	1,600	175	-	-	5	97
								4	2,500	138	-	-	57	53
								1	3,000	60	-	-	15	71
							1	5,200	75	-	-	8	88	
53,71	320	9/12	--	-	control	0	--	4	--	275	-	-	240	-
23,51	320	9/13	1900	1.4	30	7.2	cottonseed oil	5	200	81	19	77	4	71
								3	1,050	367	117	68	40	53
								2	2,500	93	63	32	18	61
							2	4,200	750	375	50	150	45	
53,71	320	9/13	--	-	control	0	--	4	--	202	-	-	148	-

<sup>a</sup>Comparison not made with control fields.

492 oil and undiluted, due to crystallization caused by solvent evaporation. The trials reported here employed an experimental 1.4 oil soluble concentrate provided by Chem-

agro Corporation. No plugging was encountered with this formulation. However, difficulty was encountered making accurate flow rate adjustments due to its high viscosity.



On the morning of August 24, 5 gallons of undiluted propoxur 1.4 OSC were applied over a 2 mile run at a flow rate of 25 fluid ounces per minute, starting 7:52 a.m. The air temperature was 76°F, wind speed of 3-4 mph. The level of control increased after the first post-treatment assessment as the toxicant had time to work. Control the first 200 feet downwind from discharge increased from 14% approximately 30 minutes post treatment to 72% at 90 minutes post, remaining at 72%, 2½ hours after treatment. Control at the 0.5 mile distance shifted during the same period from 0, to 47% and 48%, whereas little effect was seen at the 1 mile distance 0, 1% and 34% reduction observed during the same periods post treatment. The reduced flight activity during the time of day this treatment was made undoubtedly had a negative influence on the level of control which resulted.

That evening an additional 5 gallons of propoxur (1.4 OSC) was applied under a 2-3 mph breeze at a flow rate of 30 fluid ounces/minute. Intending to apply the entire 5 gallons in a run along the west and north sides of fields 8, 9, and 10, half of the tank remained at the end of the run, therefore a second application was made over the same distance to discharge the entire contents. Exceptionally good

results were obtained 12 hours post treatment, with the level of control remaining steady over the mile distance. Control results were 89% at 200 feet, 90% at 1200 feet, 93% at 2,500 feet, 94% at 3,700 feet and 87% at a mile.

Three weeks later, propoxur (1.4 OSC) was again applied in an evening application. The intended delivery of the undiluted concentrate was 40-60 oz/minute, however, the delivery turned out to be 105 fluid ounces/minute in a 6 minute run. The twelve hour post-treatment results averaged 94% at 1,200 feet, 84% at 1,600 and 91% at 4,200 feet downwind.

The fourth propoxur (1.4 OSC) application was made at 45 ounces/minute on September 14 at 6:55 p.m. on fields 2, 3, and 51, half a section in area. The 12 hour post-treatment data indicated good control out to the last station sampled. Control averaged 95% at 200 feet, 99% at 1,100 feet, 80% at 2,500 feet downwind and 83% at 4,200 feet distance (Table 4).

A comparison of the propoxur (1.4 OSC) treatments clearly indicates the evening applications were superior to the morning treatment, possibly for the reasons already mentioned. The most effective treatment was the double back application which averaged 91% reduction over an

Table 4.—Effectiveness of propoxur (OSC 1.4) applied<sup>a</sup> by cold fogger against adults of the pasture mosquito *Aedes nigromaculis* (Rancho Santa Maria, Kern, Tulare, Kings Counties, California, 1972).

Field	Acres	Date	Time	Rate fl oz/min	Sta- tions	Dist. from disch. (ft)	Pre- treat	Mean number landing/host (hours post treatment)									
								0-1		1-2		2-3		10-12		12-15	
								No.	%R	No.	%R	No.	%R	No.	%R	No.	%R
4,5,6	500		0752														
7,8,9,10		8/24	0810	25	4	200	111	83	14	22	72	24	72	95	57	-	-
					4	2,500	153	177	0	58	47	63	48	195	36	-	-
					4	5,200	118	133	0	84	1	61	34	265	0	-	-
53,71	320	-	-	0(control)	4	--	56	49	-	40	-	44	-	112	-	-	-
4,5,6			2007														
7,8,9,10	500	8/24 <sup>b</sup>	2020	30	15	200	144	-	-	-	-	-	-	15	89	-	-
					3	1,200	75	-	-	-	-	-	-	7	90	-	-
					6	2,500	482	-	-	-	-	-	-	32	93	-	-
					3	3,700	683	-	-	-	-	-	-	40	94	-	-
					6	5,200	162	-	-	-	-	-	-	21	87	-	-
53,71	-	-	-	0(control)	8	--	112	-	-	-	-	-	-	110	-	-	-
4,5,6,7																	
8,9,10	500	9/13	1900	105	3	200	1	-	-	-	-	-	-	-	-	-	-
					3	1,200	383	-	-	-	-	-	-	18	94	-	-
					2	1,600	142	-	-	-	-	-	-	17	84	-	-
					2	4,200	150	-	-	-	-	-	-	10	91	-	-
53,71	320	-	-	0(control)	14	--	202	-	-	-	-	-	-	148	-	-	-
2,3,			1855														
51	320	9/14	1920	35	5	200	131	-	-	-	-	-	-	6	95	3	96
					2	1,100	138	-	-	-	-	-	-	8	94	14	84
					4	2,500	269	-	-	-	-	-	-	48	80	53	69
					2	4,200	500	-	-	-	-	-	-	75		108	
4,7,8	-	-	-	0(control)	7	--	118	-	-	-	-	-	-	107	83	76	-

<sup>a</sup>A 2-mile run was made in each experiment.

<sup>b</sup>Double-back application.

assessed distance of one mile. The 105 ounce/minute flow rate did not prove more effective, as one might expect. Although no data are available, indications of particle drop out (ground wetting upon operating the machine in one location) were observed at high flow rates with this viscous concentrate. The lower flow rate in the double back application may have permitted production of optimum particle size thus reducing drop out, in addition to having sufficient material left for a second application.

Primiphos methyl (5 lb/gal ULV & EC) was applied in a 3-4 mph breeze on the evening of August 27 in 492 oil carrier, at 70 fluid ounce/minute. Under good weather conditions, irregular results were experienced at distances of greater than 200 feet from discharge. Control levels were 91% at 200 feet, 35% at 1,200 feet, 72% at 2,500 feet and 47% at 3,700 feet downwind (Table 5).

Dursban® (4 EC) was diluted with 492 oil to a 1 lb/gal fogging solution and applied against *Culex* adults 30 minutes after sundown on August 30, 1972. The flow rate was 56 fluid ounces/minute. Wind drift 0-1 mph, relative humidity 80%, with a high cloud cover from a southern California tropical storm. Pre-treatment counts per square yard were taken in natural resting areas around buildings before 8:00 a.m. on the morning of treatment, 12 hour post-treatment counts were taken similarly the following morning. Results out to 2,600 feet from discharge showed above 90% reduction occurred at all but two stations which were on the downwind side of buildings in areas protected by vegetation (Table 6). An average of 93% reduction was achieved in two similar locations lacking vegetation.

Of the four adulticides used in tests against OP resistant *Aedes*, propoxur was consistently more active at distances greater than 200 feet from discharge. The 10% pyrethrum, SBP-1382 7.2%, propoxur and primiphos methyl application results were essentially the same at the 200 foot level. The adults found during morning post-treatments counts at this distance can be attributed, in part, to infiltration from across the road of travel during application.

Available information on propoxur (Georghiou et al. 1966, Georghiou 1970) suggests caution in the extensive use of this material in area-wide mosquito control operations due to evidence that resistance to this material may induce tolerance to structurally related compounds in the future. Unfortunately, with the exception of the use of dichlorvos under limited conditions, propoxur is the most effective adulticide against resistant mosquitoes in California.

Operational use of non-thermal aerosols against post emergent *Aedes* populations in areas located some distance apart poses logistical problems, due to the brevity of the twilight treatment period. Contiguous large acreage treatments for control of *Aedes* adults appear feasible, and are tentatively planned for 1973 on Rancho Santa Maria. However, due to the resistance selecting potential of the sub-lethal dosages which would be received by the large number

Table 5.—Effectiveness of primiphos-methyl (Actellic 5 lb/gal, ULV formulation) applied by cold fogger against adults of the pasture mosquito *Aedes nigromaculis* (Rancho Santa Maria, 1972)<sup>a</sup>.

Field	Acres	Volume fl oz/min	No. stations	Dist. from Disch.(ft)	Mean no. land/host (hrs post)		
					Pre- treat.	10-12 No.	%
2,3,51	320	70	5	200	445	33	91
				1,200	434	442	35
				2,500	700	170	72
				3,700	700	320	47
53,71	320	0(control)	7		643	555	-

<sup>a</sup>Treatment made on September 27 at 1852. A 0.75-mile run was made. The 5 lb/gal ULV formulation was diluted with 492 process oil (Texaco Co.) to yield a formulation containing 20% of Actellic.

Table 6.—Effectiveness of Dursban® (4 EC) applied by cold fogger against *Culex* adults in a semi-urban agricultural area south of Bakersfield, California, 1972<sup>a</sup>.

Acres	Volume fl oz/min	No. Stations	Dist. from Disch.(ft)	Mean no. rest/yard <sup>2</sup> (hrs post)		
				Pre-treat	10-12 No.	%
125	56	1	800	40	3	93
			1,300	141	17	88
			1,600	15	1	93
			2,600	300	25	92
20	0 (control)	2		75	75	-

<sup>a</sup>Treatment begun on August 30, 1972 at 1010. A .75 mile run was made along a raised canal bank .25 mile upwind from most stations. The 4 lb/gal EC formulation was diluted with 492 Process oil (Texaco Co.) to make a 1 lb/gal fogging solution.

of OP resistant adults from the drift of repeated treatment, material choice and treatment pressure will require judicious attention, as annual flights of adults from this area (Smith et al. 1956) would facilitate dispersal of acquired resistance over the remainder of the southern San Joaquin Valley.

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# EVALUATION OF LOW VOLUME NONTHERMAL AEROSOLS FOR MOSQUITO CONTROL IN CALIFORNIA

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During the past two decades, mosquito control in California has de-emphasized adulticiding in favor of the organophosphorus larvicides. Recently, development of broad-spectrum insecticide resistance by larvae of two major species and recognition of the present high costs of larviciding to control mosquito larvae in rice fields have led to renewed interest in adulticiding as an operational technique.

Brown and Mulhern in 1954 reported on the performance of several thermal aerosol machines in use in California at that time and pointed out limitations and operational problems. Nonthermal aerosol generators developed for military use (Morrill et al. 1955) were later modified to apply very low volumes of insecticide and diluent (Mount et al. 1968). The wide swaths achievable on a test line basis were demonstrated in California (Stains et al. 1969; Gillies et al. 1972). There are now on the market a number of low volume nonthermal aerosol generators, and several units have been constructed and used operationally by mosquito control agencies. Personnel of the Bureau of Vector Control and Solid Waste Management have monitored and evaluated operational and test applications. This paper reports the results of selected applications on caged and field mosquito populations.

**METHODS AND MATERIALS.**—Most of the aerosol units were constructed by local mosquito control agencies. A unit from the Disease Vector Ecology and Control Center, U. S. Naval Air Station, Alameda, (Stains et al. 1969) was utilized in one test reported upon here. The control-agency machines consisted of a four-cylinder gasoline engine of approximately 20 hp direct-coupled to a positive displacement air pump. Commercially available air-shear nozzles (Afa Corp.) were modified by reducing the liquid orifice to 1/32 in with brass tubing inserts. Liquid was supplied to the nozzle by pressurizing the insecticide tank with air bled from the blower. The machines were mounted on utility vehicles.

Several insecticides and formulations were used in the tests. Malathion, registered for nonthermal aerosols and used throughout the United States, was not included in our work because of widespread mosquito resistance in California. Several tests were made with synergized pyrethrins, also registered for nonthermal aerosols. Experimental applications were made of two materials not registered for the purpose, chlorpyrifos (Dursban®) and propoxur (Baygon®).

Vegetable oils, mineral oils, and a polypropylene glycol were used as diluents when necessary. They were chosen for compatibility with the chemical being tested and for relatively low volatility.

Evaluation was based on observations made on caged mosquitoes, landing rate counts, or both. Light trap counts were made on two occasions. Caged mosquitoes used for the tests were known-susceptible *Culex pipiens quinquefasciatus* from our colony or from the University of California Mosquito Research Laboratory colony at Fresno. The cages were, for the most part, disposable paper and nylon net units described by Townzen and Natvig (1973). Landing rate observations were utilized when field populations of diurnal mosquitoes were to be treated, and were made prior to and after treatment. CDC traps baited with carbon dioxide were used to measure field populations of *Culex tarsalis* and *Anopheles freeborni*. Applications were generally timed to coincide with mosquito activity peaks.

Tests were made in two situations. One, exemplified here by the control demonstrations at the Burney Basin and Tulare Mosquito Abatement Districts, utilized open pastureland. The second, illustrated by the tests in the Colusa and Tehama County Mosquito Abatement Districts, were in residential communities characterized by such obstructions as dwellings, trees, and shrubbery.

**DESCRIPTION AND RESULTS OF INDIVIDUAL TESTS.**—Burney Basin Mosquito Abatement District.—The test area was a ranch in an isolated mountain valley. Approximately 5,000 acres are under irrigation, and much of the area produces tremendous numbers of *Aedes melanimon*, *A. nigromaculis* and smaller numbers of other species. The test layout and conditions are presented in Figure 1. The test application was made between 1900 and 2100 hours on July 25, 1972. The machine used was one built and operated by the Colusa District, described in detail by Whitesell (1973). A thermal-fog formulation of chlorpyrifos was diluted to 2 lb/gal active ingredient with the mineral oil Sontex® 75-T. A total of 12 gal or 24 lb active ingredient was dispersed during 110 minutes application time. Mosquito populations at the time of the test were moderate. Field population estimates were made at each station by three individuals who entered the field, waited about one minute, then counted the mosquitoes on one pantleg per man. The pretreatment count was made the day of treatment and the post treatment count 24 hours later, or about 15 hours post treatment. Caged *C. p. quinquefasciatus* were located at the positions shown on the map. Two hours after treatment was completed the cages were collected and replaced with new ones which were left in the field for the rest of the night.

Tables 1-3 present the results observed against field and caged populations. The average reduction in the field population 15 hours after treatment was 90%. Mortality in the cages exposed at the time of application reached 100% with-

Table 1.—Landing counts of adult *Aedes* spp. observed prior to and after a chlorpyrifos nonthermal aerosol application July 25, 1972, Burney Basin Mosquito Abatement District. Counts are totaled from those of three individual observers.

Station	Number per leg		Reduction
	Pre	Post	
1	27	0	100
2	3	0	100
3	0	0	--
4	73	5	93
5	20	6	70
6	100	9	91
7	23	7	69
8	67	11	83
9	39	6	84
10	26	7	73
11	46	1	97
12	167	5	97
13	11	8	27
14	18	12	33
15	23	4	82
16	22	3	86
17	31	4	87
18	26	0	100
19	43	0	100
20	44	0	100
21	72	1	98
22	34	3	91
23	34	1	97
24	8	0	100
25	2	0	100
26	10	0	100
TOTAL	969	93	90

in 12 hours at all stations except those at the extreme southeastern exit of the valley approximately 1½ miles from the path of the fogger. It is difficult to see any clear relationship between mortality in the first and second series of cages, but it is apparent that insecticidal activity was still present in the valley for some period of time during the night.

Tulare Mosquito Abatement District.—The Tulare District is in the area of the southern San Joaquin Valley suffering from the highest organophosphorus resistance in *A. nigromaculis* and *C. tarsalis*. Tests were conducted against *A. nigromaculis* to evaluate two adulticides, synergized pyrethrin and propoxur, in two pasture complexes of about 1,000 acres each (Figures 2 and 3). The mosquito population was evaluated by landing rate observations. Moderate populations were present. An attempt was made to use caged mosquitoes, but a change in wind direction rendered them useless. Generators used were the Colusa machine and the U. S. Navy model. The former applied 5.5 gal of a pyrethrin formulation prepared by combining equal volumes of a 10% pyrethrin/50% piperonyl butoxide formulation and

Table 2.—Mortality of caged adult *Culex pipiens quinquefasciatus* following a chlorpyrifos nonthermal aerosol application July 25, 1972, Burney Basin Mosquito Abatement District. Cages placed at stations within 1 hour pre-treatment, mortality determined at intervals post treatment.

Station	Percent mortality at hours post treatment			
	2	5	12	24
1	100			
2	7	100		
3	0	100		
4	0	100		
5	destroyed by cattle			
6	0	11	72	94
7	0	35	100	
8	0	0	16	21
9	0	0	44	56
10	0	5	68	68
11	0	87	100	
12	33	100		
13	0	72	100	
14	0	0	100	
15	6	50	100	
16	77	100		
17	75	100		
18	83	100		
19	11	100		
20	100			
21	0	8	100	
22	0	94	100	
23	13	100		
24	0	100		
25	100			
26	32	100		
Check				
1	0	0		
2	0	0	0	6
3	0	0	0	6

the solvent Klearol®, which was dispensed during 35 minutes application time. The latter applied about 4 gal of 1.5 lb/gal propoxur spray concentrate during approximately 10 minutes application time. Applications were made nearly simultaneously beginning at approximately 2000 hours on July 10, 1972.

Observations made at about 15 hours post treatment showed that the mosquito population had been markedly reduced (Tables 4, 5). The pyrethrin apparently was highly effective for ½ mile and less effective for an additional ½ mile. The propoxur application gave almost complete control for ½ mile and greatly reduced the population for an additional ½ mile. The counts for both areas remained low during observations for three days following applications.

Table 3.—Mortality of caged adult *Culex pipiens quinquefasciatus* following a chlorpyrifos nonthermal aerosol application July 25, 1972, Burney Basin Mosquito Abatement District. Cages placed at stations at 2120 - 2230 following treatment at approximately 1900 - 2100. Mortality determined 1100 - 1200 July 26, 1972.

Station	Percent Mortality
1	100
2	100
3	71
4	17
5	destroyed by cattle
6	75
7	57
8	100 (probably an error - cage may have leaked)
9	48
10	25
11	71
12	69
13	31
14	14 (ants in cage)
15	0
16	11 (ants seen capturing live mosquitoes)
17	22
18	50 (ants in cage)
19	71
20	100
21	destroyed by cattle
22	0 (ants seen capturing live mosquitoes)
23	93
24	80
25	100
26	100
Check	
1	5
2	5

Table 4.—Landing rate counts of *Aedes nigromaculis* prior to and after nonthermal aerosol pyrethrin application, July 10, 1972, Tulare Mosquito Abatement District.

Station	Pantleg counts at days post treatment			
	Pretreatment	1	2	3
1	20	<1	10	8
2	5-10	1-3	3	3
3	30-50	0	0	0
4	5-10	5-10	0	0
5	5-10	5-10	0	2

Tehama County Mosquito Abatement District.—The small community of Gerber (population 775) was treated on two occasions. The objective was to evaluate the ability of a nonthermal aerosol to penetrate a residential area. The village is well laid out for such a test, with residential streets dividing it into blocks of about 330 x 495 feet. The machine used was one constructed by the Tehama District and is nearly identical to the Colusa unit. A thermal-fog formulation of chlorpyrifos was diluted to 2 lb/gal active ingredient with Sontex 75-T.

The first application was made beginning at 2000 hours on August 21, 1972. A single pass was made with the cold-fogger traveling east along the levee road immediately south of Gerber (Figure 4). The treatment time was 11.5 minutes at a discharge rate of 27.7 fl oz/min. The second application began at 1940 hours on September 12, 1972. Single passes were made on three north-south streets at the edge of and in town (Figure 5). The treatment time was 16 minutes at a discharge rate of 24 fl oz/min.

Caged *C. p. quinquefasciatus* were used on both occasions. During the first test they were located entirely in the open and distributed throughout the village. For the second test they were distributed throughout, but were paired at each station, with one cage in the open and the other protected in shrubbery (Figure 6). At the second application, an attempt was made to measure the natural population with light traps but a rainstorm negated the results.



Fig. 6.—Placement of exposed and protected bioassay cages during nonthermal aerosol evaluations, 1972.

Table 5.—Landing rate counts of *Aedes nigromaculis* prior to and after nonthermal aerosol propoxur application, July 10, 1972, Tulare Mosquito Abatement District.

Station	Pantleg counts at days post treatment			
	Pretreatment	1	2	3
1	50	0	0	0
2	>200	<1	5-10	2
3	20->100	0	0	2
4	15-20	5-10	5-10	<1

Table 6.—Mortality of caged adult *Culex pipiens quinquefasciatus* following a chlorpyrifos nonthermal aerosol application August 21, 1972, Gerber, Tehama County Mosquito Abatement District.

Station	Percent mortality at hours post treatment			
	3	12	20	38
1	100			
2	100			
3	100			
4	100			
5	ants			
6	100			
7	68	100		
8	100			
9	100			
10	100			
11	100			
12	30	100		
13	62	85	100	
14	65	82	100	
15	100			
16	100			
17	100			
18	100			
19	100			
20	100			
21	100			
22	100			
23	22	55	66	77
24	100			
25	8	67	92	92
26	50	90	100	
27	7	57	79	86
28	54	100		
Check				
1	0	0	0	0
2	00	0	0	0
3	0	0	0	0

Observed mortalities in the caged mosquitoes are presented in Tables 6 and 7. In the first test mortality in most of the cages located in town (Stations 1-2) reached 100% within 3 hours and was total at all town stations within 20 hours. Varying insecticidal activity was noted at Stations 22 - 28, up to 1¼ miles downwind from the path of the fogger. In the second test, the application was highly effective on mosquitoes in the exposed cages. Mortality was 100% at 20 hours at all except 2 stations, which were assumed to be on the fringes of the treated area. Excellent mortality was obtained in the protected cages up to 350 ft from the path of the machine. A marked reduction in mortality was evident at greater distances.

Colusa Mosquito Abatement District.—The City of Colusa (population 3825) is adjacent to thousands of acres of rice

Table 7.—Mortality of caged *Culex pipiens quinquefasciatus* following a chlorpyrifos nonthermal aerosol application September 12, 1972, Gerber, Tehama County Mosquito Abatement District.

Station	Percent mortality at hours post treatment							
	Exposed cage				Protected cage			
	12	20	27	37	12	20	27	37
1	100				75	100		
2	100				100			
3	100				77	100		
4	92	100			64	100		
5	88	100			76	100		
6	100				100			
7	70	100			0	8	8	0
8	100				43	57	64	71
9	55	82	82	100	0	22	43	64
10	0	33	42	42	0	0	14	36
11	100				100			
12	100				100			
13	100				100			
14	100				100			
15	93	100						
Check								
1	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0

which provide a constant source of *C. tarsalis* and *A. freeborni*. Three applications were made in the populated area, but only the first is reported upon in detail here. Test conditions and layout are shown in Figure 7.

The application was made August 29, 1972, using two identical Colusa machines dispensing a pyrethrin formulation prepared by diluting 10% pyrethrin/50% piperonyl butoxide with Sontex 75-T to 3% pyrethrin/15% piperonyl butoxide. The application was made using passes at 660 ft intervals by traveling on alternate streets. Treatment began at 2000 hours and was completed at 2130 hours. A total of 13.25 gal of the diluted material was applied.

Evaluation was by adult mosquito bioassay using exposed and protected cages and with CDC light traps baited with carbon dioxide. The results are summarized in Table 8. The light trap data show that the reduction in the in-town population was unsatisfactory. The bioassay confirmed that little success was achieved in killing mosquitoes, particularly those in the protected cages.

On September 6, 1972, a portion of Colusa was treated with synergized pyrethrin diluted to 5% pyrethrin/25% piperonyl butoxide. Caged mosquitoes were used in the evaluation. At 4 hours post treatment, 100% mortality had occurred in the exposed cages at 9 of 14 stations, with slightly less overall mortality in the protected cages. A treatment was made to the same area on September 7, 1972,

Table 8.—Light trap results and mortality of caged adult *Culex pipiens quinquefasciatus* following a pyrethrin non-thermal aerosol application August 29, 1972, Colusa, Colusa Mosquito Abatement District.

Station	Total females trapped <sup>a</sup>		Percent mortality at 24 hrs	
	1 night pre-treat.	1 night post-treat.	exposed cage	protected cage
1	50	6	32	0
2	no trap		57	23
3	395	330	88	0
4	287	141	89	9
5	14	3	6	85
6	40	37	0	0
7	no trap		75	15
8	123	277	11	91
9	no trap		67	2
10	47	24	0	0
11	186	102	8	3
Check				
12	175	185	no cages	
13 <sup>b</sup>	no trap		23	
14	3847	3255	no cages	

<sup>a</sup>Mainly *Culex tarsalis*, *Anopheles freeborni*, *Aedes melanimon*.

<sup>b</sup>May have been in treated area.

using chlorpyrifos diluted to 1 lb/gal active ingredient by combining equal volumes of Dursban ULV and the polypropylene glycol P-400®. Caged mosquitoes were placed at 23 test locations. Mortalities were generally greater in the exposed than in the protected cages, but an unexplained control mortality obfuscated the results.

**DISCUSSION AND CONCLUSIONS.**—The experience gained in conducting these tests has shown us that the low volume nonthermal aerosol generator can be a useful tool in mosquito control programs. Several mosquito abatement districts have constructed units in their own shops which are capable of producing a highly effective aerosol, with which rapid and effective coverage of large areas can be achieved. Test results also indicate that it may be possible to obtain penetration of shrubbery and other obstacles in residential areas to reach resting mosquitoes.

Application and evaluation problems encountered were similar to those with other adulticiding methods. Field effectiveness is difficult to evaluate against other than diurnal species actively seeking blood. Meteorological conditions, particularly wind, are critical. Mosquito insecticide resistance overrides any hoped-for advantages in efficacy of this type of aerosol (Gillies et al. 1972).

There are several factors related to nonthermal aerosol applications about which more information is needed. A major one is the droplet size spectrum being produced by each of the various machines as compared with a yet-to-be-defined optimum in the several situations to be found in California. It is not yet clear which chemicals, formulations, and concentrations should be selected for each situation encountered. More work is needed to determine the best application time as related to meteorological conditions and mosquito activity. Potential environmental hazards, if any, from materials applied by this technique must be understood in order to answer questions concerning the possible effects of toxicants within and drifting beyond the target area.

#### Acknowledgments

Appreciation is extended to Peter B. Ghormley, Manager, Burney Basin Mosquito Abatement District; Dennis J. Ramke, Manager, Tulare Mosquito Abatement District; Melvin L. Oldham, Manager, Tehama County Mosquito Abatement District; and Kenneth G. Whitesell, Manager, Colusa Mosquito Abatement District for their participation in the tests.

Insecticides for the tests were furnished through the courtesy of the Dow Chemical Company, the Chemagro Corporation, and the McLaughlin Gormley King Company. The Thiokol Corporation loaned an Imp for the Burney Basin test, and the Disease Vector Ecology and Control Center, Naval Air Station, Alameda, provided a nonthermal aerosol generator and a vehicle for the tests in the Tulare District. Light trap results for the Colusa District test were provided by staff of the Sacramento Area Office, Bureau of Vector Control and Solid Waste Management.

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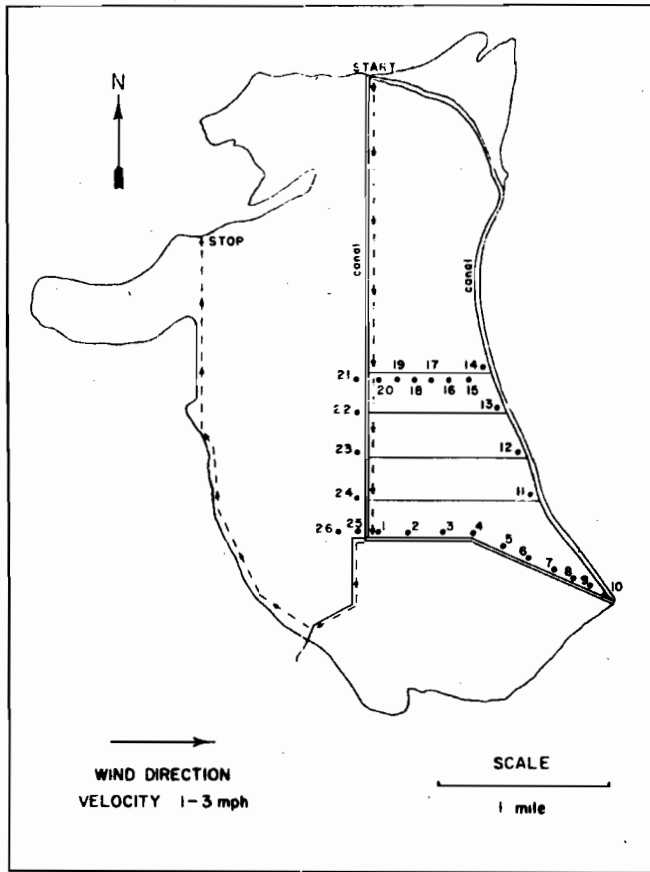


Fig. 1.—Goose Valley, Burney Basin MAD. Nonthermal aerosol application of chlorpyrifos, July 25, 1972.

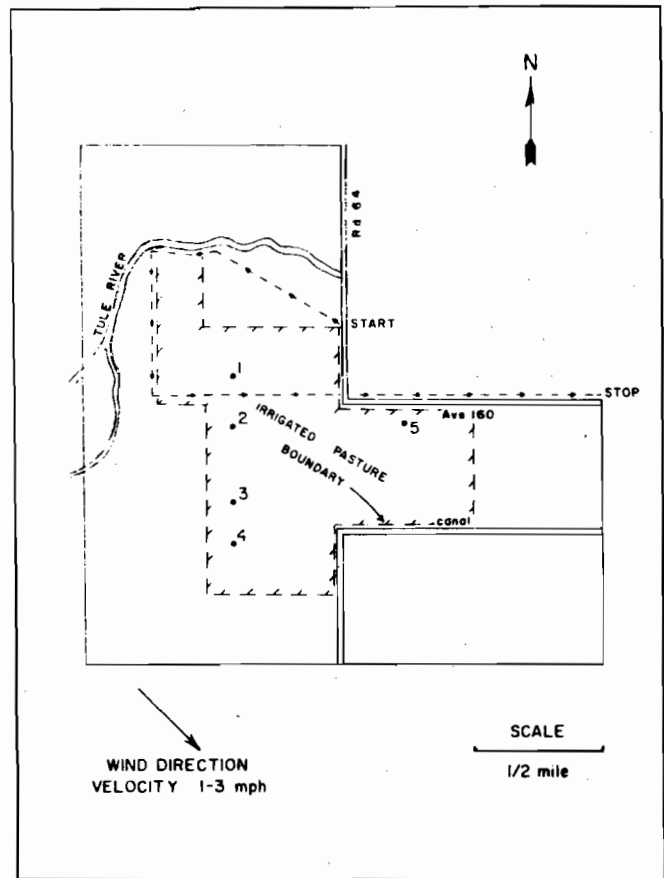


Fig. 2.—Culp Pasture, Tulare County MAD. Nonthermal aerosol application of pyrethrin, July 10, 1972.

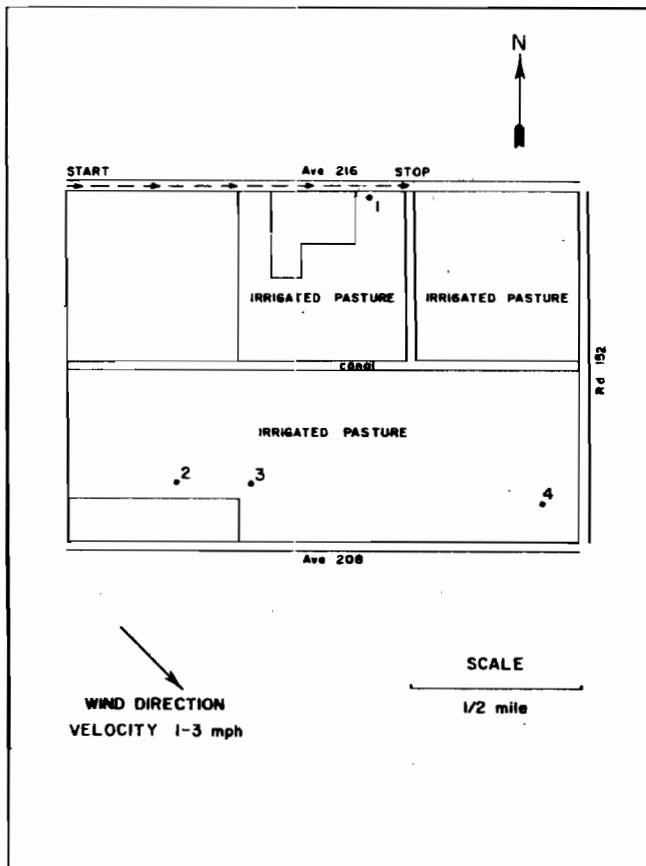


Fig. 3.—Faria Pasture, Tulare County MAD. Nonthermal aerosol application of propoxur, July 10, 1972.

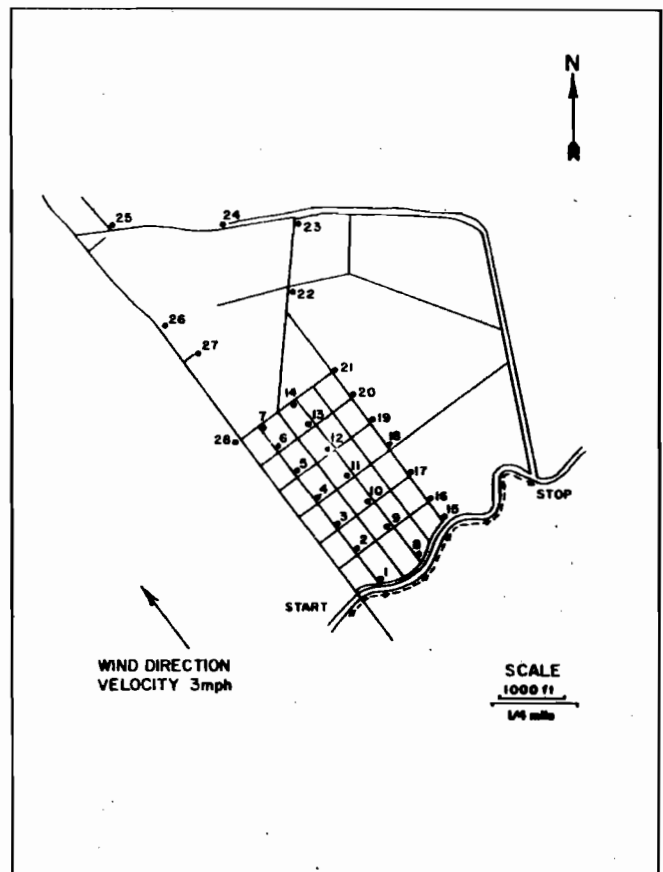


Fig. 4.—Gerber, Tehama County MAD. Nonthermal aerosol application of chlorpyrifos, August 21, 1972.

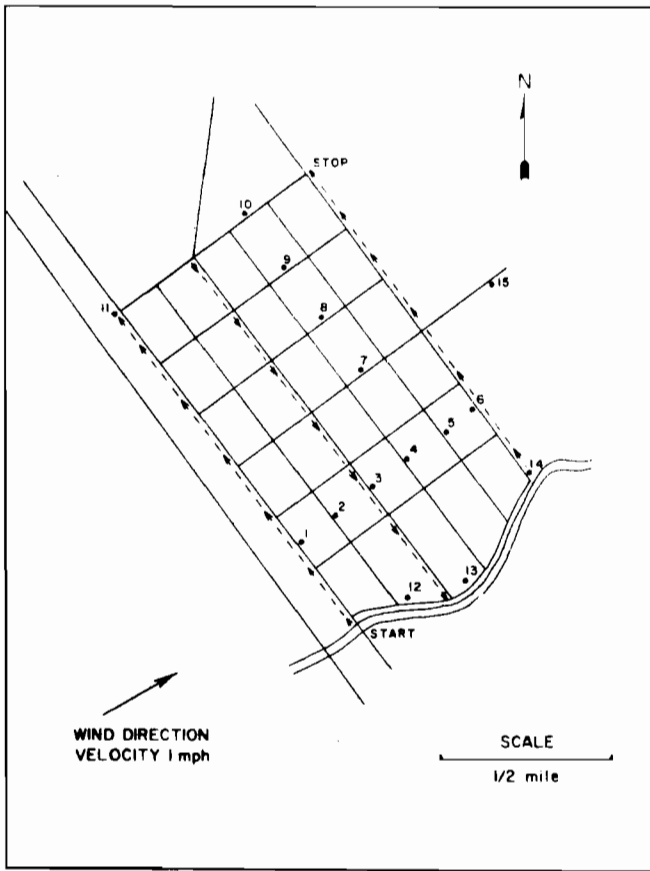


Fig. 7.—Colusa, Colusa County MAD. Nonthermal aerosol application of pyrethrin, August 8, 1972.

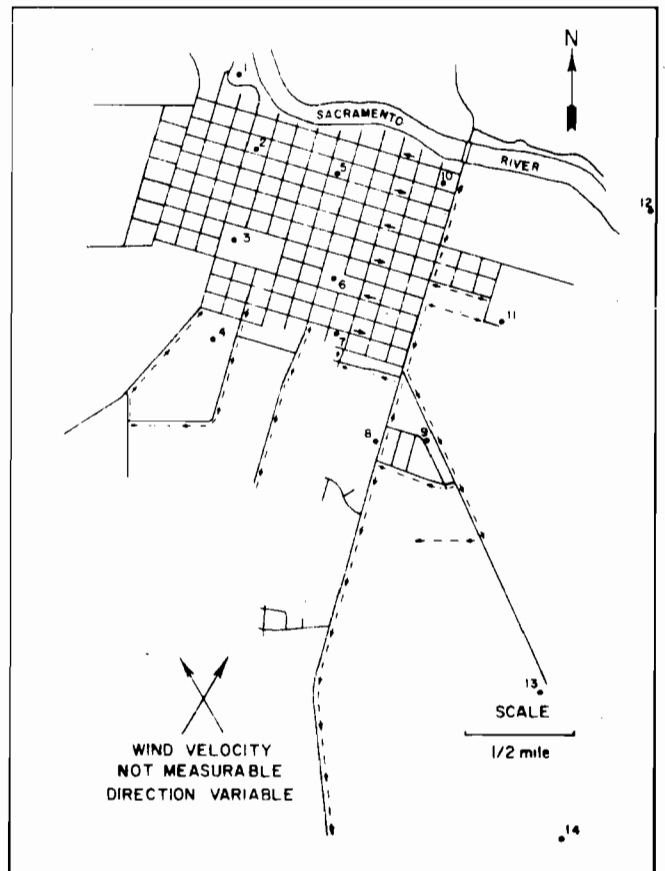


Fig. 5.—Gerber, Tehama County MAD. Nonthermal aerosol application of chlorpyrifos, September 12, 1972.

# THE THIRD ERA OF MOSQUITO CONTROL IN CALIFORNIA HAS JUST BEGUN

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Mosquito control in California has passed through two distinct eras, which in retrospect might be called the "First Era of Integrated Control" and the "Miracle Insecticide Era". It appears that a new "Era of Comprehensive Control" is just beginning.

In the first or pre-DDT era good work was done with tools and materials of limited capability. Some integrated control was practiced, with ditching to facilitate the free movement of the predatory mosquito fishes in the salt marshes, some attention was paid to drainage in the uplands, and judicious use was made of oils and Paris green larvicides. Because the costs were high and resources were limited progress could be made only on limited fronts, but the people received protection from mosquito-borne diseases, notably malaria, and from intolerable mosquito annoyance.

The miracle insecticide era began with DDT, followed by various other organochlorine and organophosphorus and carbamate insecticides. Immediate results were obtained, and the program was popular with the people, who therefore were willing to invest in mosquito control agencies. Over a period of nearly a quarter of a century the local agencies have accumulated excellent facilities including trained and experienced personnel, good equipment, knowledge of the terrain and of the problems, operational facilities and funds, and knowledge of other approaches. Therefore, we can better meet the more difficult problems ahead. We may have been too preoccupied with pesticides, but the net total effect which was obtained can only be assessed as very good!

The "Era of Comprehensive Mosquito Control" has already begun but we are only in the transition stage. Local agencies are approaching comprehensive mosquito control, which will involve naturalistic control, inclusive of but not limited to biological control; physical control including mechanical elimination of breeding places or "source reduction"; the restricted use of chemical pesticides; and various measures designed to influence landholders to prevent mosquito production on their lands.

Table 1 is a graphic presentation of the range of activities which may occupy a local agency engaged in "comprehensive control".

Mosquito control is now more than 70 years old. The first large scale effective public-supported program began in 1901 in Havana, Cuba. Intensive operations were undertaken which effectively controlled *Aedes aegypti*, the vector of yellow fever, resulting in practical control of the disease. The program was expanded to include malaria control. Elimination of breeding places was featured, supplemented by temporary treatment with petroleum oils. Col. W. C. Gorgas, who was in charge, said: "I suppose that in the year after the discovery by the Army Medical Board that the mosquito was a means of spreading yellow fever, we spent \$100,000 outside of the routine sanitary work on mosquito extermination." Of course, the expenditure of \$100,000 in

1901 in Havana would be equivalent to more than a million dollars in California in 1973. Colonel Gorgas went on to report that in addition to effectively controlling yellow fever, malaria was reduced from 325 deaths in 1900 to only 39 deaths in 1903. He added that "the consolidated report of January, 1901, before mosquito control work began, showed 26,000 water deposits producing mosquito larvae within the city limits and the same report for the following January showed less than 300."

Significant features of the work in Havana, and the more extensive program in Panama which later cleared the way for construction of the Panama Canal, were intensive initial surveys to locate breeding places, followed by equally intensive continuing surveillance to guide control operations. The title "integrated control" had not been coined, but this program integrated elimination of breeding places with temporary control, mechanical protection by screening, and public education. There was a strong legal control feature by which persons who persistently maintained mosquito sources were fined or subjected to other legal action.

The impact of the successful programs in Havana and in Panama was felt strongly in the United States and within a few years, mosquito control programs began in California, New York and New Jersey. For many years thereafter, the Cuba and Panama programs were cited as examples to prove that mosquito control could be successful.

In the first era of mosquito control, the workers had mainly oil as a larvicide, some information about the individual mosquito species being attacked, and imaginative approaches which were not limited by preconceived ideas or by demonstrated dramatic reductions obtained through the use of highly effective chemicals. Previous to these successful demonstrations, people in many parts of the country where mosquitoes were numerous considered the control of mosquitoes a hopeless task. It was common for citizens to remark: "there have always been mosquitoes and there always will be, mosquito control is hopeless".

**THE FIRST ERA OF INTEGRATED CONTROL.**—From 1900 to 1945, entomologists, engineers, program managers, and field workers all contributed a vast amount of information about ways and means of controlling mosquitoes and of eliminating the breeding places. Good progress was made but was not uniform and notable setbacks occurred on both the east coast and the west coast, where efforts to control mosquitoes were too limited in area to be successful. However, enough success was obtained to encourage communities and agencies.

Outstanding reports of studies appeared during this era: a report in 1904 by John B. Smith on the mosquitoes of New Jersey; the book by Dr. William B. Herms and Harold F. Gray, which appeared in California in 1942; and the compilation of 57 papers edited by Dr. Mark O. Boyd which appeared in 1949. These three extraordinary works (and

Table 1.--Elements of comprehensive mosquito control performed by mosquito control agencies.

Comprehensive Control Inclusive of all known control methodology as applicable Integrated Control	<p><b>A. Biological Control (a major element of Naturalistic Control)</b></p> <p>Predators, principally fish and aquatic insects          Parasites          Pathogens          Plant inhibitors of mosquito production</p> <p>Abiotic factors (inanimate natural forces) which may affect the mosquitoes, the biological enemies or the sources.</p> <table style="width: 100%; border: none;"> <tr> <td>Natural runoff</td> <td>Alkalinity</td> </tr> <tr> <td>Percolation</td> <td>Salinity</td> </tr> <tr> <td>Evaporation</td> <td>Acidity</td> </tr> <tr> <td>Transpiration</td> <td>Sunlight</td> </tr> <tr> <td>Turbulence, wave action, currents</td> <td>Shade</td> </tr> </table>	Natural runoff	Alkalinity	Percolation	Salinity	Evaporation	Acidity	Transpiration	Sunlight	Turbulence, wave action, currents	Shade	<b>NATURALISTIC CONTROL</b> Employs various biological & physical factors similar to those found in nature		
	Natural runoff	Alkalinity												
Percolation	Salinity													
Evaporation	Acidity													
Transpiration	Sunlight													
Turbulence, wave action, currents	Shade													
<p><b>B. Physical Control (Source Reduction) - elimination or manipulation of the breeding places</b></p> <table style="width: 100%; border: none;"> <tr> <td>Water management</td> <td>Land preparation and management</td> </tr> <tr> <td>  Regulation</td> <td>  Filling</td> </tr> <tr> <td>  Circulation</td> <td>  Grading</td> </tr> <tr> <td>  Drainage</td> <td>  Draining</td> </tr> <tr> <td>  Reuse</td> <td>  Crop selection and management</td> </tr> <tr> <td>  Impoundment</td> <td>  Weed control</td> </tr> </table>	Water management	Land preparation and management	Regulation	Filling	Circulation	Grading	Drainage	Draining	Reuse	Crop selection and management	Impoundment	Weed control		
Water management	Land preparation and management													
Regulation	Filling													
Circulation	Grading													
Drainage	Draining													
Reuse	Crop selection and management													
Impoundment	Weed control													
	<p><b>C. Chemical Control</b></p> <p>Ovicides (not usually practicable)          Larvicides (smaller areas treated to control mosquitoes which affect larger areas)          Pupicides (infrequently employed)          Adulticides (particularly useful in emergencies and where larval resistance to chemicals occurs)          Repellents          Weed Control          Growth regulators          Attractants</p>	<b>TEMPORARY CONTROL</b>												
	<p><b>D. Mechanical Barriers</b></p> <p>Screening          Screening of buildings          Use of temporary barriers as bed nets, mosquito-proof clothing</p>													
	<p><b>E. Landholder Motivation to Cooperation</b></p> <p>Public information and education          Individual persuasion and cooperation          Legal action and enforcement          Interagency cooperation</p>													

others published during the pre-DDT period) had several things in common: each gave full acknowledgment to the fact that natural enemies and other natural factors provide adequate control in only a limited number of situations; and each presented the mosquito problem as a complex combination of natural and man-made situations, not susceptible to control by a simplistic program, but which

should be attacked by a combination of preventive and corrective measures. Each proposed that mosquito control programs employ the natural enemies, by modifying the environment from one favorable to mosquito production to one which would favor the natural enemies and inhibit mosquito production, and by utilizing chemicals as a temporary supplement to naturalistic control and source re-

duction.

These authors showed that there is no fully satisfactory system or pattern by which control measures can be classified. One arbitrary classification system attempts to include all control measures in categories entitled "biological control", "physical control", and "chemical control". This is a convenient classification if one is willing to accept the fact that there is considerable overlap. For example, when one employs chemical control the major expense and effort involved often is the physical and mechanical application of the chemical. Furthermore, the chemical is effective because it disrupts the biological life processes of the mosquito. Thus, the operation might be called either physical, mechanical, biological, or chemical. However, through common usage, we have all come to know that when we say "chemical control" we mean the control of mosquitoes by the application of chemicals, and use of the term creates no barrier to communication.

When we refer to "physical control", which in California is often called "source reduction", the message may not be so clear because some workers like to include in "source reduction" chemical control measures, biological control measures, and such physical control measures as ditching, filling, diking, etc. Others regard "physical control" or "source reduction" as being limited to the making of a physical change in the environment, as by ditching, which removes the water in which mosquitoes occur.

Likewise, there is considerable confusion relating to the definition of biological control. One group includes only the adding of living natural enemies which will attack the mosquito either in the larval or adult stages. Other workers would like to include any operation which makes only a minimal change in the habitat but which allows natural limiting factors to function more effectively. This may more properly be called "naturalistic control" since it is an extension of what occurs in nature and includes both biological and physical factors. An example of this has been the evolution of the program which prevents emergence of mosquitoes from the salt marshes which are open to tidal action. Here ditches are installed to connect from the places which hold water to the natural waterways, not for draining the water completely, but to allow complete circulation of the tide waters, bringing mosquito-eating fish to all portions.

THE MIRACLE PESTICIDE ERA.—We may pass over this quarter of a century of progress rather lightly, not because it was unimportant but because we lived through it and are familiar with its impressive array of accomplishments. An incomplete outline of the highlights of accomplishment may be presented as follows:

1. An extraordinary level of protection from mosquitoes was provided for nearly 25 years.
2. In 1952 the span of one encephalitis outbreak was shortened, and steps were taken to avert epidemics on 3 other occasions.
3. Substantial contributions were made to the economic development of the state:
  - Realty values were safeguarded.
  - Laborers in the outdoors were protected from intolerable annoyance.
  - People using recreation areas received protection from

mosquitoes.

- Cattle in protected areas were protected and could gain more weight.
4. Great advances were made in equipment and methods for the dispersal and utilization of pesticides, mainly by aircraft, resulting in greater efficacy and economy.
  5. The environment for man and for other useful animals was provided protection in at least 3 ways:
    - Primary benefits accrued by preventing pollution of the air space by pest and vector mosquitoes.
    - Secondary protection was accomplished by limiting insecticidal applications to only the breeding places, instead of broadcast adulticiding, and by applying only minimal selective amounts. Persistent pesticides were largely eliminated long before required by legislation and regulations.
    - Biological control was substituted for pesticides in many situations. *Gambusia affinis* was introduced in 1922 and has been distributed from the Oregon line to the Mexican border, reducing mosquito production and serving to augment the food chain of bass and other sport fish.
    - Biological control by native coastal fishes has been extended by ditching on the salt marshes to aid circulation of the water on and off the marshes with the tides.

THE BEGINNING "ERA OF COMPREHENSIVE CONTROL".—Good progress already has been made by local agencies in assessing their individual needs and restructuring programs accordingly. Problems vary and within a "comprehensive control" framework the solutions will be equally variable.

An impressive armamentarium of technology is available. The problem is a matter of which technology to integrate into a program welding into one whole various elements of biological control, physical control, and chemical control to form one practical, functional unit.

Technologies which may be applied include:

- A. Survey: A resurvey of the problems and a census of the breeding areas.
- B. Biological control: Mosquito control agencies are in tune with the environment, and are quite aware that beneficial natural control forces are constantly functioning but usually require manipulation if they are to be effective.

Examples include:

1. In residential areas garden pools must be kept free of excessive vegetation if fish are to be effective.
2. In agricultural areas reservoirs and ditches must have clean banks and be free of aquatic weeds; fish must be added in pump and sump recirculation systems.
3. The industrial mosquito sources such as log ponds have been kept mosquito free by management which prevents excessive pollution (cold decking of logs instead of floating) so that fish can function.
4. Natural sources (salt marshes) are kept mosquito free by ditching to circulate tide water bringing in native fish; or by impounding so that salt is reduced and mosquito fish (*Gambusia*) can function.

Natural enemies usually function together; therefore, where fish and predaceous insects occur together, they must be compatible in numbers and behavior.

Beyond the predators are the parasites and the pathogens -- current research is now illuminating the part they play. Also, the manipulation of genetics appears to offer considerable promise.

#### C. Physical control:

1. Elimination of standing water by filling, grading, ditching, and pumping offers a useful simplistic approach in some situations, but will not provide a satisfactory solution to the more complex problems where water must be retained on the land for agriculture, wildlife or other uses.
  2. Where water must be conserved, design can be incorporated in the impoundments and in the water conveyance systems which will minimize mosquito production.
  3. Ground water recharge basins are a special type of impoundment, subject to well known standards for mosquito control.
  4. Where habitats for marsh wildlife are maintained mosquito control becomes much more complex, demanding proper land layout and preparation, water management of the highest precision, biological control, and in some cases the judicious use of selective pesticides. Each situation is individual, requiring surveillance and joint action by wildlife management and mosquito control specialists.
5. Irrigated agriculture continues to be the most extensive source of mosquito problems. The mosquitoes of irrigation water habitats have a rapid life cycle and develop in huge numbers where irrigation results in water standing on the soil long enough to allow the development and emergence of mosquitoes. Ultimately this problem must be met by the landholders who control the irrigation water and who therefore have legal responsibility to avoid the creation of a public nuisance.
  6. The public mosquito control agency evidently must accept a more dynamic role in expanded surveillance to keep constantly informed of the status of mosquito sources; it must determine why sources exist where they do and who is responsible; it must inform the responsible landholders and help them to find ways to attain their land-use objectives without creating a mosquito nuisance; it must find ways to obtain the active cooperation of all other agencies and groups who have a vested interest in the land or water where the problems develop; and it must continue to seek other more effective and economical solutions to the developing problems which for the future appear to be almost insurmountable.
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# PAST, PRESENT AND PREDICTABLE FUTURE OF AIRCRAFT IN MOSQUITO CONTROL

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Mr. Mulhern has proposed that this panel review the history of mosquito control to glean from the past what we may use in the future. Panel members represent many years of mosquito control experience, about 110 years. To put it another way, with due respect to my colleagues, we are a group of old timers. Most of us have been battling the mosquito for 25 years and we will be stepping aside within a few years. This period may become recognized in history as the era of chemical control.

DDT, then known as the "miracle insecticide", became available in 1946. Prior to that time mosquito control in California was limited mainly to source reduction, biological control through the use of *Gambusia*, and chemical control with oil larvicides. None of these measures were too highly developed nor completely effective.

Compared to the sophisticated programs of the present the scope of mosquito control was quite limited. There were about 18 to 20 districts, in most instances providing mosquito control in and about the cities or urban areas. Most districts ranged in size from 12 to 40 square miles. With limited resources and ammunition with which to do battle, some control was achieved, but all too often its effect was negated by mosquitoes migrating into the small areas under control.

There were no flood control dams on the Kern River in 1946 and one could count on extensive annual flooding of overflow lands from east of Bakersfield to 20 miles west. Dipping at the edge of the flood area yielded *Aedes* larvae numbering about 500 per dip. It was apparent that the ground equipment control measures then in use were not going to get the job done. We were also confronted with another mosquito source of equal magnitude — the annual flooding of thousands of acres of unlevelled native pasture lands scattered through a major portion of the district. It appeared that the only possible solution to these two problems would demand the use of aircraft.

During the spring months we nibbled away at control over acreage we could reach with ground equipment. The job was of such magnitude our efforts were of little value in controlling the populations of emerging *Aedes*. In surveying the possibility of using aircraft to control the large acreages I went to the local agricultural aircraft operators, including a branch of Atwood Crop Dusters, then the largest in the state. I wanted to know if they could successfully spray a solution of DDT by air over the areas of mosquito production. Aircraft operations in California were limited to the application of dusts and there were no aircraft available for spraying liquids. When I asked about installing a liquid spray system on at least one plane the response was surprising. The manager of the operation wanted to know: "What would have to be done?" I suggested that a system would probably consist of a tank to hold the spray material, a pump and a boom with spray nozzles attached.

Again the response was disappointing: they would only attempt such a novel idea if I could guarantee that it would work! That was impossible as I didn't know it would be successful. Faced with an impasse on aerial spraying, we hired a commercial operator at \$1.00 an acre to apply DDT dust. A standard dust formulation was applied at about 2 lbs per acre, until we could obtain a suitable wettable powder formulation for subsequent use.

Results were encouraging and reinforced my conviction that liquid sprays should be tried. Diligent search for some system of spray equipment suitable for use on an airplane continued to be frustrating, until shortly before the season ended we found what we were looking for. A pilot in Reedly had a crude spray system already installed on a plane. The owner-operator of a machine shop there had assembled a liquid spray system and was trying to get the commercial operators interested in aerial spraying.

We hired the plane and pilot for the last few weeks of the 1946 season. Results were so gratifying that a decision was made to purchase and operate our own aircraft. During the winter of 1946 two military surplus Stearmans were purchased and equipped for spray operations. The spray equipment installed consisted of tank, wind driven pump and spray nozzles on a boom. After several years of trying pumps and nozzles we settled on the use of wind driven gear pumps using nozzles with number 1 discs and as many as 40 nozzles on the boom.

At first oil solutions were applied at a rate of 2 gallons per acre. Extensive field testing subsequently showed that aqueous solutions were as good or better than the oil solutions. We then switched operationally to the aqueous formulations and gradually reduced the volume applied to ½ gallon per acre and concurrently increased the size of nozzle orifices to number 4.

During the late 40's and early 50's agricultural aircraft operators and other mosquito control agencies began developing and using aerial spray equipment. Using aircraft for spraying of the new organic pesticides we succeeded in attaining a level of mosquito suppression which previously had not been possible. The attack was primarily against larvae but adults in the fields treated were killed also.

New districts were formed and old ones expanded in size. These factors contributed to more effective mosquito control and more extensive use of aircraft. By the 1960's aircraft for mosquito control had become so commonplace that little thought was given to the effectiveness of aircraft. Their use has provided capability to do the most effective and economical job possible. At the period of maximum use 13 local mosquito control agencies were operating 24 fixed wing and rotary wing aircraft, and 10 agencies were contracting for additional commercial planes. Acreages treated annually amounted to about 1,500,000 acres with about 90% treated with district

equipment and 10% under contract. The development and use of ULV spray equipment made aircraft spraying even more effective and efficient.

Resistance to insecticides coupled with increasing restrictions upon the use of aircraft and sprays has resulted in a significant decrease in the use of aircraft for mosquito control in California during the past year or two. We can expect to see this trend continue for the next few years but in time I believe the trend will reverse as research and development now under way produces more effective oils, newer toxicants, and biological agents which will again enable us to provide a desirable level of mosquito control.

The airplane can be expected to remain the most effective and efficient piece of equipment for the application of such control agents.

Rotary and fixed wing aircraft for aerial inspection and photography have proved invaluable in flood years. In 1969 rapid inspection of vast flooded acreages via helicopter provided districts with up-to-date information on which to act, proving the value of rotary wing aircraft in disaster situations. With greater emphasis on source reduction, aerial surveys and photographic documentation of the need for measures have been useful in convincing landholders of the need to correct problem areas.

## EVOLVING ECOLOGICAL AWARENESS AS A BASIS FOR MEETING THE MORE DIFFICULT PROBLEMS OF THE FUTURE

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For many years ecology was a required course for any college student majoring in biology. Very often the course was dry and uninteresting and followed a dictionary definition: Ecology . . . dealing with the mutual relations between organisms and their environment . . .

In recent years, with the development of the faddist type of "ecologist", the term has about as many definitions as there are people that use it. To a great majority of these individuals it means the maintenance of an absolute and unchanging condition called "natural balance".

I prefer to define ecology as "the dynamic interaction of all natural systems, organic and inorganic, living and dead".

I would emphasize the word dynamic, since all things on earth — whether biologic or geologic — are in a constant state of evolution and change. It would be a pretty dull place in which to live if this were not so.

All organisms use the ecological system for their own sustenance whether it be the simple absorption of inorganic matter from the soil and air, or feeding on other organisms.

Nature is also an organizer, starting with the complicated chemical systems and simple single celled organisms to which life has been given, and evolving them into complex systems of specialized cells which we call higher plants and animals.

Many living organisms are also organizers. These organizations range from the honey bees and termites, to group-living higher animals or complexes such as our human society. All lead to the same end result: the benefit and protection of the group.

Man's special place in the scheme of nature is due to his increasing capacity to reason and invent. Unfortunately, it seems that his ability to protect himself from adversities in his environment and to invent tools for his existence, protection, and convenience seem to have outstripped his ability to reason the consequences of his inventive genius.

At one time, man was a hunter-gatherer with a simple family or pack society and had little more impact on the environment than any other omnivorous animal. Mosquito control consisted of slapping and brushing motions. Then he learned to make fire and found that if he threw green vegetation on the fire to make more smoke, this helped protect him from the little pests.

Eventually he discovered that those biting things developed from little wigglers and tumblers in the water and also learned that certain diseases were transmitted by mosquitoes, which changed them from a simple pest to a public health menace.

About the same time, man found out that he could grow food and fiber more efficiently if he irrigated it. This had the effect of growing mosquitoes more efficiently, too.

As mosquito control evolved man learned that he could plant *Gambusia affinis* in mosquito sources and they would eat the larvae and pupae; biological control was born! Man also found that if he put oil or other chemicals on or in water, or on adults, he could kill mosquitoes easier. Then the great chemical revolution was on.

Public Health became Public Health and Comfort. The people demanded more and more comfort and the fastest and easiest way to provide it was through chemical control. The public agencies formed to protect the people from mosquitoes were under the gun. Of course, biological and mechanical control such as drainage were not discontinued, but they were put on the back shelf because results were slower and not as spectacular.

Evolution continued to march on. Came the time of ecological awareness; it was found that:

1. Insecticidal chemicals had undesirable side effects and were often misused;
2. Draining natural wetlands had undesirable side effects on



desirable animals;

3. Drainage from agricultural land polluted natural waters and that was undesirable.
4. Mosquitoes could develop a resistance to insecticides faster than new ones were being developed.
5. Our old friends, *Gambusia affinis* were believed to be damaging to some natural inhabitants of waters into which they were introduced and that was not desirable.

Granted that we saw much of this problem developing –

Granted that we have been trying for years to obtain funds for increased research –

Granted that we have increased our efforts to educate the public as to their responsibility in mosquito prevention –

Granted that we have tried to increase source reduction operations in one way or another –

Granted we have recognized that our efforts do have an impact on the environment –

Granted that much of the hysteria over ecology is calming down and that more rational attitudes are being expressed – nevertheless, ecology is here to stay. It has had an impact on organized mosquito control and will continue to do so.

The greatest impact on mosquito control so far has been in slowing down the development of new insecticides. This has been felt especially because of our excessive dependence on chemical control.

Mosquito control workers in California have always attempted to select insecticides carefully and to use them with as much precision as possible. This effort will need to be strengthened and encouragement given to the development of even more specific insecticides. The insect development inhibitors show promise.

Dependence on insecticides must be reduced. Newer and more specific materials will be more expensive and reduced use will decrease the pressure which leads to the development of resistance.

The effects of environmental requirements on source reduction activities have not yet been much felt, but I predict that they will be. Already it has been said that excess or waste agricultural waters will have to be retained on the

property. How this will be reflected in ground water pollution, I don't know. I think more tail water return systems are in the offing in place of off-farm drainage. Drainage into canal systems for reuse down stream may also become more prevalent.

Requirements set to reduce water pollution could have the effect of forcing a reduction in water usage. This would be advantageous to mosquito control and should be encouraged.

The Association and its members should be more active in keeping their needs and problems before the water quality control authorities as they are considering the disposal of agricultural effluents. Mosquito control could be either helped or hurt by requirements promulgated for water quality control.

Research in naturalistic control should be increased wherever and whenever possible. Developments in this area are of the greatest importance – especially in the control of mosquitoes being produced in intermittent waters. For the short run, I believe they will be more efficient. However, people may have to live with a few more mosquitoes.

Water users must be made to accept the fact that they are responsible for mosquito production. As the pressures of the environmental protection movement increase, and they will, the success of mosquito control will depend more and more on changing the water wasting habits of people.

I foresee the ultimate integration of mosquito control through chemical, naturalistic, source reduction, and public education measures. Each of these techniques should modify the others to form a more cohesive and effective whole.

Finally, the concerns for a better environment are here to stay. They will probably lead to aggravation in more forms and reports to make out. They must be kept in mind and allowed for as programs change in the future and they may be responsible for some program modifications. Concerns for the environment can be a help as well as a hinderance in mosquito control and must be kept in mind. Advantage should be taken of the environmental protection programs where they can be of assistance.

Above all, mosquito control programs must be as flexible as possible to benefit from new technology and to adapt to the continuing evolution of the human society which will undoubtedly bring pressures and demands which cannot now be foreseen.

# A CHANGING EMPHASIS OF MOSQUITO CONTROL IN CALIFORNIA: SURVEY, PLANNING, PERSUASION, NEGOTIATION, COOPERATION AND LEGAL ACTION

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The management of all mosquito control programs in California has used and is using the basic approaches needed to accomplish the economic control of mosquito breeding.

These basic approaches, as listed by the title for my part of this panel, are survey, planning, persuasion, negotiation, cooperation and legal action. The degree of emphasis by any one of these six basic approaches may vary from one program to the next depending on the particular objectives of each program as well as on the type of community involved — whether agricultural, industrial, urban, recreational or a combination of each type.

The future emphasis on the control of mosquito breeding will have to be intensified and will have to include all six of the basic approaches. In addition, the conventional methods of executing these six basic approaches will have to become more sophisticated and more complicated and more time consuming.

By the conventional method, I mean the direct approach by management to the person, corporation or public agency directly responsible for the prevention, abatement or control of a mosquito breeding nuisance. This direct contact by only two concerned parties, the property owner and the district, permits a simple and quick solution to the problem.

The need for a change in emphasis is already in the making by the public's demand for the protection, preservation, and development of our dwindling natural resources. The public's decision to regulate the use of these natural resources was recorded by the passage of the California Coast Line Conservation Initiative, better known as Proposition 20. The organization of the six Regional Coast Line Conservation Commissions and the State Conservation Commission is currently under way. The recent California Supreme Court ruling in the "Friends of Mammoth Decision" has already unleashed a flood of Environmental Impact Statements (EIS) in conformance with the California Environmental Quality Act of 1970. These statements are required for major new construction, including drainage systems and flood channels, water conservation and spreading projects, housing developments, recreational areas and other similar projects which create, either directly or indirectly, mosquito problems.

The management of a mosquito control program will have to expand the conventional concept of "survey" from that of physically locating newly created breeding sources to that of locating and appraising potential breeding sources that may be created by a proposed project or development.

The time and place for effective "planning, persuasion, and negotiations" will have to be changed from our direct contact with the property owner after the problem has been

created to the hearing rooms of planning commissions and other regulative bodies prior to the final design and approval of the proposed project or development.

Although cooperation with the property owner or public agency has traditionally been the cardinal approach by mosquito control management, future solutions to mosquito breeding problems will require even more exacting cooperation by the district, even to the extent of discontinuing effective and economical physical methods, and implementing naturalistic and biological methods requiring a higher degree of technical supervision and maintenance. This type of situation has already occurred in Orange County with the abandonment of the proposed design for a conventional concrete lined flood channel in a narrow right-of-way and the adoption of a "natural" drain-way in a wide, undisturbed natural right-of-way.

The public demand for protecting, preserving and developing ecological areas for fish and wildlife is already reflected in current and planned projects for green belts, open space, national parks, recreational lakes and water ways. The public awareness of the environment and its demand for ecological areas are a blessing in disguise for mosquito control programs throughout California. The control of mosquito breeding requires the awareness, cooperation and support by the general public. Public support has always been strongly evident in the past and can be anticipated in the future since the policy of mosquito control in California is complementary with that of the environmental conservationist. This mosquito control policy, referred to as "comprehensive mosquito control" has been defined by Thomas D. Mulhern in the opening paragraph of his article, "An Approach to Comprehensive Mosquito Control", as published in California Vector Views, Vol. 19, No. 9, September, 1972. I believe it is appropriate to review Mr. Mulhern's definition at this time.

"Comprehensive mosquito control may be defined as scientifically planned, continuing, carefully executed programs aimed at insuring the effective control of mosquitoes of all species which may bring disease or harassment to man and domestic animals. Within this concept all of the known technology is to be applied when appropriate; but preventive measures are to be emphasized, principally naturalistic control and source reduction. A fundamental element of this philosophy is that chemical larviciding and adulticiding are to be considered interim measures, to be used only until basic and permanent control is achieved, or where problems of small magnitude occur so infrequently that environmental management is not justified, or where the danger of epidemic disease demands immediate action."

The changing emphasis of mosquito control in California places additional requirements for manpower and technical skills on mosquito control management. As environmental regulations are implemented, and as demands for ecological conservation areas increase with urbanization, present day management staffs will not be able to adequately "survey" potential mosquito breeding sources or to effect preventive control measures by "planning" and "persuasion" and "negotiation" with those agencies and commissions responsible for establishing standards and for approving new projects and developments. In addition, the control of mosquito

breeding under these environmental conditions will not only be more time consuming but will require a higher degree of technical skill and precision than that presently required.

It is my conclusion that each Board of Trustees should appraise the impact of this changing emphasis of mosquito control within its district and anticipate the time when additional manpower needs and technical skills will be required. With the current public support for environmental protection, preservation, and development, a preventive mosquito control program can help meet the objectives of the district at the most economical cost.

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## ADVANCING MOSQUITO CONTROL IN DIFFICULT TIMES

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A trustee of a mosquito abatement district is unique in that he represents the public as well as the landowner. The emphasis now being placed on enforcement of law by some districts as a means of abatement is somehow in my mind a solution of last resort. Our role should be one of fostering and aiding cooperation; not just cooperation between the board and the manager, but cooperation between the public, the landowner and the district.

Education is undoubtedly the best tool that we can use and you have all been deluged with methods and means of accomplishing these things. In your efforts at education, however, you are given a priority in the minds of the public. That priority is established by the surroundings. When an individual is being plagued by mosquitoes you may have his first priority at that moment, but in the off season – now – where do you stand?

The so-called environmentalist has caused a revolution to occur and you and I are only a small part of that resolution. Air pollution, water pollution and land use are the broad spectrum in which he works. Let's look at some of the results.

In order to attain the air pollution standards now set, you buy a new car with pollution abatement equipment that reduces mileage, performance and just plain driveability of the car. Yet, the people in charge say, that in order to meet the clean air standards set in the State of California, we will need to remove 86% of the cars from the road within the time span of the next five years.

The California Water Quality Control Board has set standards for the sanitation districts that the Chairman of the C.W.Q.C. Board, Win Adams, stated flatly he realizes are impossible to meet. He stated, in an address made on September 13, 1972, that the industry and Water Pollution Control Agency should accept a bad law in exchange for the promise of benevolent administration. Cost in the County of Los Angeles alone for capital expenditures will be 470 million dollars in five years with an annual upkeep of something like 35 million in 1972 dollars.

Land use and other environmental considerations have brought on an energy crisis nation wide. In the area served by the Southern California Gas Company, in December, 1972, there were 11 days when gas was completely shut off for all industry and electric power generators on interruptible service. For 20 days of the 31 days in December, 1972, there was a partial shut off. Of the 1907 manufacturers

using propane, some 30 were in real trouble and curtailed or shut down production completely and sent employees home with unemployment insurance their only income. I can find no one who will tell me the number of employees out of work for this cause, but let me assure you, the number is going to increase.

When people tell you to increase your public relations budget and get your program across to not only the public but your legislators, congressmen and senators, you're competing against more than just the apathy of the "boob tube". You're competing against the worst and the best. The worst is the person who says "don't bother me with the facts, I've already made up my mind," and the best is the one who looks at your problem and tries to fit it into the perspective of the complicated structure in which we have enslaved ourselves.

Another matter that has developed since the charge given to the Committee of the Future has been the recent "Dream of Governor Reagan". Governor Reagan has translated his dream into action by appointing Lieutenant Governor Reinecke in charge of a task force to streamline local government. His charge to the task force is to determine ways and means of reducing the tax burden of local taxpayers by elimination of inefficient local governments and particularly to eliminate or reduce the so-called multiplicity of special districts. The task force has assigned the job of fact gathering to the Council on Inter-governmental Relations. This Council has had hearings and has gathered information from the counties through the Supervisor's Associations; the cities through the League of California Cities, and from special districts through the offices of the California Special Districts' Association.

It appears to me that these various special interests (regional government adherents) may have pre-conceived notions that mosquito abatement districts, other districts, and many cities are no good; that they do not perform legitimate functions and should go, regardless of the facts. It would appear to me that all trustees of mosquito and pest abatement districts should make it their business to look into this subject and then make their voices heard.

All I can say at this point is "Good Luck and VAYA CON DIOS."

# A UNIFYING FRAMEWORK FOR MOSQUITO CONTROL

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Probably one of the most startling discoveries we all make once we have been in mosquito control for a while is the tremendous breadth of the field. One moment we need to be an engineer, the next minute an administrator, public relations man, or entomologist. It makes for interesting work, but it does create at least one problem: we must depend upon a diversity of disciplines to provide us with guidelines for our activities. The pitfall is that a discipline, be it engineering, biology, chemistry, or management science, has facts, theories, approaches and languages that are specifically applicable to its own particular field. It can be very frustrating to attempt to develop a cohesive mosquito control program while borrowing from these specialized disciplines.

The "systems" approach appears to have some promise in overcoming the above problems. Frank Stead introduced the subject to mosquito control in 1966 at one of our California Mosquito Control Association Conferences. It offers hope of providing a common perspective, and an interdisciplinary language, with which we can approach our mosquito control activities. A selected bibliography follows which I hope may be of some value to those wishing to further pursue the subject:

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# THE K-COEFFICIENT: A MEASURE OF ASSOCIATION BETWEEN MOSQUITO LARVAL SPECIES IN BREEDING SOURCES

Iver E. Bradley<sup>1</sup> and Jay E. Graham<sup>2</sup>

Since 1956, and in conjunction with the control activities, the South Salt Lake County Mosquito Abatement District has been conducting research on larval populations and their ecology. As a part of this program, when an inspector finds a positive source or pool (one with mosquito larvae present), in addition to such data as number per dip, temperature of the water, size and depth of the pool, etc., he collects a vial of representative larvae, with a particular effort to get a sample of all species present. These sample vials are brought into the laboratory and the species identified and recorded. There are five major species, *Aedes dorsalis*, *Culex tarsalis*, *Culiseta inornata*, *Culex pipiens*, and *Aedes vexans*. Only 1% of the positive sources are without one or more of these species.

One of the interests of the research and control programs is related to the questions of when and why these several larval species occur together and to develop some measure of the degrees of association or lack thereof.

Earlier we (Graham and Bradley 1962) found that a particular species increased in abundance when it was able to occupy a larval habitat without other species being present. Over a period of years there has been considerable fluctuation in the ability of species to occupy larval habitats alone and also considerable fluctuation in the degree of association between species in the larval habitat.

Early in the season (April and May) positive larval habitats are usually relatively few and most contain *A. dorsalis*; *C. inornata* is also well represented. During June, July, and August, positive larval habitats become more numerous; *C. tarsalis*, *C. pipiens*, and *A. vexans* appear with high frequency along with the two early species. All of the five major species were found on some occasions with each of the other four. None of our species is ubiquitous during this period. It is during this time of greatest larval production that a measure of association is very useful and meaningful.

Before proceeding with the discussion of our measure of association, we define the following symbols:

- (1)  $N$  = total number of positive sources (pools) for the time period of consideration.
- (2)  $N_x$  = number of pools with larval species,  $x$ .
- (3)  $N_{xy}$  = number of pools with both larval species,  $x$  and  $y$ .
- (4)  $P_x = \frac{N_x}{N}$ , the proportion of pools with larval species  $x$ ; the simple probability of  $x$ .

- (5)  $P_{xy} = \frac{N_{xy}}{N}$ , the proportion of pools with both larval species,  $x$  and  $y$ ; the joint probability of  $x$  and  $y$ .
- (6)  $Q_x = 1 - P_x$ , the simple probability of not- $x$ .

It is rare that  $P_x > 0.50$  and, so far, it has always been the case that  $P_x < 0.60$ . The normal range for  $P_x$  is between 0.10 and 0.45, and we have no cases where  $P_x + P_y > 1$ .

Certain properties of a measure of association are desirable:

- 1) If the positive sources of two species were independent of each other, this measure should be zero. Statistical independence is defined mathematically in the usual way: two species are said to be independent in their choice of larval sources if and only if  $P_{xy} = P_x P_y$ . Two larval species are independent if the proportion of pools containing species  $x$  in the subset of pools showing species  $y$  is the same as the proportion of pools containing  $x$  to the totality of pools.
- 2) If the two species have a tendency towards dissociation, the measure should be negative. The extreme case of this negative dependence would be that where no pool contains both species, i.e., the sources would be mutually exclusive. For this extreme case, we would like the measure to be -1.00.
- 3) If two larval species tend to occur in the same pools, the measure should be positive. The extreme positive dependence could occur if the breeding spots of one species  $y$ , were a subset of those of another species,  $x$ ; i.e.,  $P_y = P_{xy}$ . In this case, we would like the measure to be +1.00.
- 4a) It would also be desirable to know something about the distribution of this measure. Using  $K$  as a symbol for this measure, we should be able to test the null hypothesis,  $H_0: K = 0$ , against the alternative,  $H_a: K \neq 0$ .
- 4b) Beyond this, we should be able to test for significant differences between two given  $K$  values.

After giving careful consideration to several measures of association that have been used by biologists (reviewed by Southwood 1968, Wilhm 1972, and others), we decided to use the following:

- (1)  $K = \frac{P_{xy}}{P_x P_y} - 1$  if  $P_{xy} \leq P_x P_y$ .
- (2)  $K = \left( \frac{P_{xy}}{P_x P_y} - 1 \right) \frac{P_x}{Q_x}$  if  $P_{xy} > P_x P_y$  and  $P_x > P_y$ .

(See technical appendix for the relationships that exist between  $K$  and the coefficient of linear correlation and the traditional  $\chi^2$  [2 by 2] contingency test).

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For our particular situation, this measure satisfied most of the desirable criteria.

- 1) It is extremely simple; i.e., it is easy to calculate and easy to interpret.
- 2) If species  $x$  and  $y$  are statistically independent in their selection of breeding spots, i.e.,  $P_{xy} = P_x P_y$ , then  $K = 0$ .
- 3) If between two species, the breeding spots are mutually exclusive, i.e.,  $P_{xy} = 0$ , then  $K = -1$ .
- 4) If  $P_{xy} > P_x P_y$ , then, as we would like,  $K$  is positive, and if the sources of species  $y$  is a subset of those of species  $x$ ,  $K = 1.00$ .
- 5) The standard error of  $K$ ,  $\hat{\sigma}_K$ , is not as simple as we would like but is a function of  $N$ ,  $P_{xy}$  and  $P_x P_y$ . Even so, we can calculate  $\hat{\sigma}_K$  and therefore are able to test the desired hypotheses. (See technical appendix for table of  $\hat{\sigma}_K$  values and the formula for  $\hat{\sigma}_K$ .)

To illustrate the use of  $K$  we have chosen to present and compare the data and results from the two years 1970 and 1971 (June through August). See Table 1.

The total number of sources for the two years were about the same, but, as far as larval production was concerned, the two years were significantly different. In 1970, each *Aedes* species was found in about 18% of the pools and the other three species, *C. tarsalis*, *C. pipiens*, and *C. inornata* were each found in about 30% of the pools. In the same period in 1971, both *Aedes* species dropped as a percentage of total pools, *A. dorsalis* to 14% and *A. vexans* to

10%. The percentage of *C. pipiens* pools dropped from 33% to 23%. With a drop in proportion of these three species from 1970 to 1971, the other two, of course, increased rather markedly. The percentage of pools with *C. tarsalis* jumped from 33% to 57% and for *C. inornata* this percentage went from 31% to 41%.

Several questions are of interest. (1) For which of the pairwise combinations of larval species can we reject the null hypothesis? (2) What is the direction of association for these rejected? (3) Are there significant differences between  $K$  for a given pair in 1970 and the corresponding  $K$  in 1971? (4) Can these significant shifts be explained?

The species combinations in Table 1 are arranged in decreasing order of the 1970 coefficients of association ( $K$ ). It is interesting to note that with one exception, *C. pipiens* and *A. vexans*, the 1971  $K$ -coefficients were in the same order as those for 1970.

The only positive associations were between *A. dorsalis* and *A. vexans* for both years. These are both significant and the degree of association in 1971 was significantly higher than that for 1970; there was more mixing in 1971.

There were only two  $K$ 's where the null hypothesis would not be rejected and curiously enough, both of these were in 1971 and both were exactly zero (correct to two decimal places). These were for *C. tarsalis* and *C. pipiens* and for *C. inornata* and *C. pipiens*. The selection of breeding sources by these two pairs in 1971 were apparently independent.

In all the other sixteen cases the coefficients of association were significantly negative; i.e., there was a pairwise tendency for these species to seek different breeding spots. No pairs were mutually exclusive, but 12 of the  $K$ 's were between  $-0.50$  and  $-0.80$ . These 12  $K$ 's were all of the cases involving one of the *Aedes* species with one of the other three species, indicating a generally high degree of dissociation with *A. dorsalis* or *A. vexans* and any of the other species.

Without exception, the 1971  $K$ 's were at least as large as the corresponding 1970  $K$ 's, indicating a general tendency to higher association (more sharing of the same breeding spots) in 1971.

Of the ten possible combinations of the five major species taken two at a time only *A. dorsalis* and *A. vexans* showed a preference for the same type of larval habitat.

All other combinations tended to select different habitats. The tendency to select different habitats in the two years of data reported here was greatest with *C. tarsalis* and *A. vexans*, with the other possible combinations showing intermediate values between the positive  $K$  of the two *Aedes* species and the extreme negative  $K$  of *C. tarsalis* and *A. vexans*.

All of the possible combinations of two species showed a tendency to associate more in 1971 than in 1970 except *A. dorsalis* and *C. tarsalis* which remained the same, and perhaps *C. tarsalis* and *A. vexans* where the difference is not highly significant.

Further study is required to define precisely the changes in larval habitats from year to year and how weather factors influence them. The  $K$ -coefficient will be a useful tool in these studies. We now know which shifts are significant and direct our research efforts accordingly.

Table 1.— $K$ -coefficients and probabilities for June through August 1970 and 1971.

Species	1970		1971	
	$P_x$			
<i>A. dorsalis</i>	.184		.139	
<i>C. tarsalis</i>	.332		.572	
<i>C. inornata</i>	.306		.405	
<i>C. pipiens</i>	.328		.225	
<i>A. vexans</i>	.176		.099	
	$P_{xy}$	$K$	$P_{xy}$	$K$
<i>A. dorsalis, A. vexans</i>	.053	+14	.038	+28
<i>C. inornata, C. pipiens</i>	.084	-16	.091	.00
<i>C. tarsalis, C. pipiens</i>	.081	-26	.129	.00
<i>C. tarsalis, C. inornata</i>	.061	-40	.162	-30
<i>A. dorsalis, C. tarsalis</i>	.027	-56	.035	-56
<i>C. inornata, A. vexans</i>	.016	-70	.017	-58
<i>A. dorsalis, C. pipiens</i>	.017	-72	.012	-62
<i>A. dorsalis, C. inornata</i>	.016	-72	.021	-63
<i>C. tarsalis, A. vexans</i>	.012	-79	.016	-72
<i>C. pipiens, A. vexans</i>	.011	-80	.011	-50
	N = 3886		N = 4381	

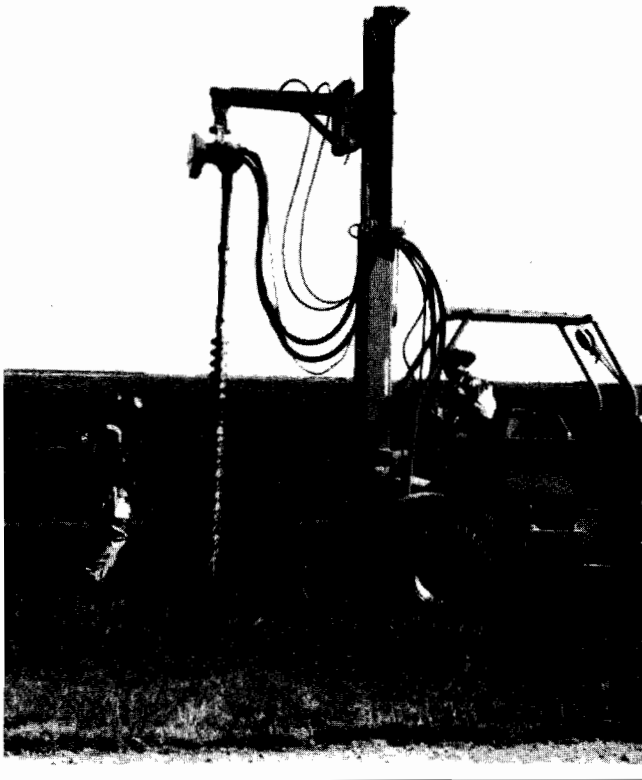


Fig. 1.—View of the boring rig being used in 1972.

When flooded, that rate proved to be undesirably high. We then bored at rates of 85 and 70 holes per acre. Eighty-five was slightly more and 70 was slightly less than necessary. In the remaining trouble spots, 75 holes per acre drained off standing water within three days. Altogether, we bored 104 holes in the second field, but only 1.2 acres were involved in mosquito production, so 90 holes (at a rate of 75/acre) would have been sufficient. In our judgment, vertical drainage was an effective and potentially cheap process for obtaining mosquito abatement in the second field.

We also bored 131 holes in a 40-acre alfalfa field at the southwestern edge of the City of Hanford. At the time the work was performed, about  $\frac{1}{4}$  of the alfalfa had been drowned out but a return system for tail water was being installed. Our work was predicated on the anticipated effectiveness of the return system. However, the grade within the field proved to be so slight (and in some checks even nonexistent) that instead of the anticipated 75 holes per acre in a total of about 1.7 acres, we ended with an average of about 39 holes per acre in 3.4 acres. Although the existing holes appeared to move water downward at a satisfactory rate per hole, additional holes were not bored because good agronomic practice indicated that the field should be regraded.

We worked also on a farm near 4th and Kent, several miles southeast of Hanford in Kings County. The 40-acre pasture apparently had an adequate grade, but the need for releveling was so great that vertical drainage appeared to be an impractical process. However, on an experimental basis, we bored an overall total of 50 holes in 3 of the 35 checks. In all three checks, we obtained adequate drainage for mosquito control at rates of 75 per acre in two checks and at slightly more than 85 in the third even though water averaged about 12 inches deep in the problem area of the third check when the flooding period ended.

We worked also on a horse farm about  $3\frac{1}{2}$  miles southeast of Clovis, California, in Fresno County. The primary problem was in the exercise lots where standing water and mud stemmed both from the sprinkling system and from natural precipitation. Water permeable strata that gave good vertical drainage were encountered near the 10-ft level.

Altogether, then, in 1972 we bored on four rather widely separated farms in Fresno and Kings counties and encountered water-permeable strata on each farm. Therefore, we have not changed our original idea that vertical drainage can be a useful and practical tool in mosquito control.

Since this is a progress report, some statement of future objectives again seems in order. First, we believe our present boring rig can be improved in efficiency to the extent that one man can bore an average of 15 to 20 holes per hour to a depth of 11 to 12 feet in terrain of average difficulty. Of course, much more basic information must be acquired before the actual potential of vertical drainage can be estimated.

#### Acknowledgments

We gratefully acknowledge the help and advice given to us by the following: T. Cheney and R. Gagliardini of Valley Foundry and Machine Works, Fresno, California; George Ogden, Jr., 4581 North Fowler Avenue, Clovis, California, who served as our consultant and guide for the original design and most subsequent modifications and who performed all the welding and machine work on the boring rig; the Consolidated MAD for personnel, a truck, and aggregate during our first two outings at the Gilbert Farm; the Kings MAD for personnel, a truck, and aggregate at the Natali Farm; members of our laboratory staff who assisted in various phases of the work, especially David G. Brandl and Terrell W. Tucker. We thank William C. Bianchi, Research Leader, USDA-ARS, Fresno Ground Water Recharge Field Station, Fresno, California, and Richard C. Husbands, Senior Vector Control Specialist, Bureau of Vector Control and Solid Waste Management, California State Department of Public Health, Fresno, California, for peer reviews of the manuscript.

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# VERTICAL DRAINAGE FOR MOSQUITO CONTROL IN 1972 — A PROGRESS REPORT

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In two previous meetings of the CMCA, we discussed the concept of a vertical drainage for mosquito control in small, problem areas in well contoured, judiciously watered irrigated fields in the alluvial soils of the Central Valley of California (Lewis & Christenson 1971, Lewis et al. 1972). At the 1972 meeting we presented evidence that such drainage — and control — had been obtained by boring holes into underlying, water-permeable strata. Most holes that drained well in 1971 ranged between 9 and 11 feet deep. We reached that depth by attaching a 5-ft-long auxiliary boom to the 9-ft-long loader bucket arms of a small skip loader to achieve an overall 14-ft-long boom to which we affixed a hydraulic boring head. But even with a 14-ft-long boom, our 9-ft and deeper holes transcribed a distinct arc.

The best solution to the problem of arch-shaped holes obviously lay in using a boring rig with a downward thrust that deviated but little from the perpendicular. But since we were unaware of a suitable machine which we could copy or acquire that would bore to those depths, a considerable portion of the time allocated to the drainage project in 1972 was concerned with the construction and ongoing improvement of such a boring rig. If the project was to be effectively continued, we considered that approach unavoidable because a practical concept of vertical drainage hinges on the availability of a maneuverable, dependable rig that will bore many holes per hour in difficult terrain at a reasonable cost per hole.

This paper is a report on the rig we constructed and some of the data obtained with it.

**MATERIALS AND METHODS.**—To a compact, 4-wheel-drive vehicle equipped with a truck engine and a power take off<sup>1</sup> we attached a 2-section forklift mast<sup>2</sup> and appropriate hydraulic equipment including a double-acting 7-ft-long stroke hydraulic cylinder within the forklift mast. The piston rod of the cylinder was attached to the inner, movable section of the mast so positive downward power could be brought to bear on that section. At the rear and near the lowest extremity of this movable section, we attached a leaf-chain roller in such a position that it straddled, and could give downward thrust to a leaf chain which had one end attached to the rear center of the fork carriage and the other end attached to the middle of the upper cross-brace of the fixed, outer

<sup>1</sup>A self-propelled power source for an electric generator used to start jet engines on military aircraft. Acquired without cost to us from military surplus. The machine had a wheelbase of 84 inches, but shorter or longer wheelbases should serve equally well.

<sup>2</sup>From a forklift with a recommended capacity of 6,000 lb. Acquired without cost to us from military surplus. A lighter, and therefore cheaper, mast should serve equally well.

portion of the mast. Thus with the double acting hydraulic cylinder, we could exert smooth, positive downward and upward force on the fork carriage throughout its 14-ft range. To obtain vertical holes, then, we mounted our hydraulic boring head on a short boom affixed to the fork carriage. The boom was hinged and braced so that it could, with the auger attached, be quickly swung to the side by means of a small double acting hydraulic cylinder. The carriage and mast could then be lowered, and the rig put in traveling position with the auger cradled along the side of the vehicle. Conversely, and just as quickly, the machine could be converted to the boring position from the traveling position.

We have bored holes to a depth of 13 feet with the outfit. On every occasion, our hydraulic system has provided sufficient downward thrust to penetrate whatever hardpan or other difficult substrata or obstruction we encountered. With an 8-ft-long auger plus extensions we commonly reach a depth of about 11 feet in from 1½ to 2 minutes in terrain of average difficulty. We might add that because of our final objective we have seldom bored in terrain that could be easily penetrated.

As for the mosquitoes in the fields where we have worked, we have yet to observe pupation of *Aedes* spp. on the third day after flooding, but we have observed pupation on the fourth day that was followed later by emergence. But we have not observed pupation followed by emergence unless standing water was present at the time of pupation. For mosquito control then, only the places that contain water on the third day after flooding need to be drained.

All holes in 1972 were bored with the new rig described by using a 6-in diameter auger (Figure 1). All holes were filled with a coarse, ¾-in aggregate as in 1971.

**RESULTS AND DISCUSSION.**—We bored in two fields about 6 miles east of the Fresno Air Terminal. On this livestock farm, the fields, with the exception of low spots that have developed, were well-levelled with a good slope, but the soil was somewhat heavy for good internal drainage.

When field number one was graded and levelled, several feet of topsoil had been moved from a knoll to lower areas. In this knoll area we encountered rock — or a rock-like formation — from about six feet to a depth beyond the 13-ft lower limits to which we bored. This cemented material would not lend itself to the process of vertical drainage. In two other troublesome spots outside the knoll area, we penetrated into a gravelly substratum at about the 9-ft level that did take water readily; but overall our process gave but little relief in field number one.

In the second field, we encountered water-permeable layers at depths from 8½ to 10½ feet. In a preliminary trial, we bored at rates of slightly more than 150 holes per acre.

# POTENTIAL VECTOR PROBLEMS ASSOCIATED WITH SUBSURFACE UTILITY ENCLOSURES

Charles M. Myers<sup>1</sup>, Ed Colson<sup>2</sup>, and Gilbert L. Challet<sup>3</sup>

It has been known for some time that various types of subsurface enclosures are capable of serving as suitable sites for mosquito breeding. Past experience has demonstrated that underground systems are also capable of serving as harborage for pest and vector species other than mosquitoes. Species included in this list are black widow spiders, several species of cockroaches, and yellowjackets. Larger enclosures may also be capable of harboring rats.

Little has been said by mosquito control agencies in the past concerning utility enclosures largely because the numbers of enclosures have been relatively small. In the last few years, however, there has been a very rapid expansion of underground electrical distribution systems. Unlike the older and more familiar large walk-in vaults, the new residential transformer enclosures are only slightly larger than the equipment they house. Access is available from the surface and the system is designed for one-man operation.

There are several different equipment designs. Basically they consist of a concrete enclosure which houses the transformers, a bolt-on open grill top, and usually a shallow ( $\pm$  6 inch) gravel base. The enclosures may be round or square. The round type, for vertical transformers, is generally 36 inches or 42 inches in diameter and about 6 to 7 feet deep. The square horizontal types are usually about 3 feet deep. This unit, being shallower, results in less contact with ground water but presents a larger surface area.

All of the transformers in these enclosures are rated for underwater service. The utilities are, however, concerned about corrosion and silting. It would, therefore, seem to be in everyone's best interest to maintain as dry an environment in the enclosures as practical.

The residential-type transformers are rated for at least 12 KV. The potential dangers inherent in having non-qualified personnel in contact with this type of equipment precludes adequate sampling by mosquito abatement or health department personnel. Mosquitoes have, however, been recovered from flooded enclosures by both the Pacific Gas and Electric Company and the Southern California Edison Company.

Initial estimates indicate at least 5 to 10% of the enclosures contain water. As many of the residential areas "mature" and more sprinklers are installed in areas where they will influence flooding of the enclosures, larger numbers of flooded enclosures might be anticipated.

Approximately 8,000 to 10,000 underground units are being added to the system per year in California. In new residential subdivisions there is one transformer for about every 10 houses. This indicates that in the future these enclosures could be as numerous as street draining catch basins. Some mosquito control districts are already involved in a surveillance program on utility enclosures. For example, Orange County MAD has over 1,000 of these enclosures under surveillance at this time.

Representatives of the Bureau of Vector Control and Solid Waste Management have corresponded with the utilities involved, principally PG&E and SCE, for over a year regarding potential vector problems in these enclosures. Both companies' staffs have been cooperative in attempting to elucidate as well as find solutions for the problem. Jointly over 14 possible means of eliminating the problem have been considered. Most of these have, unfortunately, been eliminated as feasible solutions for various technical reasons. The utilities are currently in the process of analyzing the flooding potential of their enclosures in several representative areas of the State. It is our mutual aim and desire that once we better understand the sources of flooding water, we can, by engineering, eliminate water and hence mosquitoes from these enclosures.

In the interim, the Bureau of Vector Control and Solid Waste Management is presenting the following installation guidelines to those utilities involved in underground systems:

1. In no case should the top of open enclosures be installed below grade.
2. Enclosures should not be placed in areas where high water tables exist and could cause them to flood unless the enclosures are sealed against mosquitoes.
3. Enclosures should not be installed in areas of obvious surface runoff or in areas subject to frequent overhead sprinkling unless provisions are made to prevent water from entering into or standing in the enclosures.
4. The use of increased gravel pack, dry wells, or leach lines should be considered in areas where light to moderate water encroachment is anticipated.
5. Property owners should be encouraged not to flood the enclosures.

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# WILL LOCAL GOVERNMENTS BE MERGED INTO REGIONAL GOVERNMENTS?

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In recent years in California special districts have come under the scrutiny of a "research corporation", the California Legislature, Local Agency Formation Commissions, and the County Supervisors Association of California. In Oregon, bills that would eliminate special districts have been introduced into the legislature. These have been frontal attacks, addressing themselves to possible consolidation of similar or unlike types of local governments.

Now attention is being directed toward the formation of regional governments. Governor Reagan has said that possibly some counties could be merged with other counties, cities with other cities, and that the "myriad of special districts should be joined together". He has selected Lt. Governor Ed Reinecke to head the Council on Intergovernmental Relations, and this body will hold hearings for the purpose of streamlining local government.

Mr. Reinecke has pointed to the 5,758 governmental entities in the state, a figure that includes cities, counties, school districts and special districts.

Arguments being used to support regional government are:

1. Too many special taxes;
2. The public cannot attend the Board sessions of all of the special districts at which budgets are set;
3. The County Supervisors can reduce general county taxes, but the total local tax load may not be reduced because special districts can raise theirs;
4. There is a proliferation of special districts;

5. There are too many laws and acts governing the several types of special districts;
6. The Board of Supervisors is more visible than the boards of special districts;
7. There should be more widely-based planning; and
8. Federal money is available for area-wide planning.

Arguments favoring continuation of special districts are:

1. Special districts are responsive geographically and personally;
2. Special districts are responsible to the people they serve; revenues are derived only from beneficiaries;
3. They serve well because they have a limited geographical area for performance;
4. They are performing well and their work could not be performed better by someone else;
5. Special districts are flexible and can adapt to the need for change;
6. Over-all planning should be kept separate from over-all implementation; and
7. There are steps that are short of regionalization, such as joint power authority or a uniform district act.

If changes are to be made, and that appears to be at least possible, it would seem appropriate and desirable for special districts to become acquainted with all officials involved and to work closely with them to be sure that the important charge and efficient work of the districts is recognized.

## EVOLUTION OF A MOBILE COMMUNICATION SYSTEM

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Evolution is a continuous change from a lower, simpler state to a higher, more complex state, a progressive development. The Southeast Mosquito Abatement District, in the past twenty-one years since its formation, has been undergoing such a change. Our communication system is such an example.

Twenty-one years ago we did not have direct communications with the personnel in the field; consequently, breakdowns in the field could not be serviced, in some cases, for as long as five to seven hours, thereby losing the use of that unit for a full day and sometimes for two days. Service requests could not be answered in most cases in less than twenty-four hours.

In 1958, the Southeast MAD decided to experiment with a low-cost, two-way radio system. The area to be covered at the time was approximately 175 square miles. Funds for this project were limited, and it was decided to try Citizens' Band Equipment which was not too expensive. This equipment operates as AM in the low band frequency with a total input of 5 watts. Application was made to the Federal Communications Commission to operate in this frequency. One base station plus 7 mobile units were operated the first year. The results of that first year were very beneficial to the District's operational program. Breakdowns and the resulting waits for assistance were in most instances reduced to one to two hours in contrast to the previous five to seven hours. Service requests in 50% of the cases were being answered the same day, resulting in improved public relations and quick control of local breeding sites.

Within the next two years, all District vehicles were equipped with two-way Citizens' Band radios, reducing the amount of lost operational time and greatly improving the community relations program of the District.

All good things seem to come to an end or at least become more difficult to achieve; so it was with Citizens' Band Two-Way Radio. When the general public found out that they could have two-way radios in their homes and cars at very low cost, the frequencies seemed to explode. Everyone wanted to get on the air and chit chat with their friends. The jam of people using all the frequencies at this time made

it almost impossible to communicate with our base station. Within two years, we were reduced to mobile communications within one to two miles of each other. Our lost time in the field due to breakdown and other problems was again increasing. Public relations were beginning to suffer because of our inability to answer service requests as required for best possible service.

Change from a lower, simpler state to a higher more complex state was again in order. Discussions with our supplier, Federal Communications Commission, and Frequency Coordinator determined that we should apply for a frequency in the local Government Service. The consensus in our case, because of the area to be covered, 528 square miles, was that we should remain in the low band, but at a much higher frequency, with FM radio. Application was made, and we were assigned an open frequency three years ago to operate at 180 watts with a base station tower of sixty feet. This system gives the District clear coverage of twenty-five to thirty-five miles in all directions.

Funds again became a critical item when we began to change a phase of our operations. A complete change of our communications system in a short time was not possible because of lack of funds. It was decided to take three years to make a complete change to the new system. The cost of a 40 watt, tube-type transceiver for a mobile unit was about 4 times greater than a 5 watt C.B. transceiver. The cost of a similar solid state unit was about 10 times the cost of a C.B. unit. The tube type was selected to begin our changeover to Local Government frequency and in the process, our supplier agreed to accept our C.B. units as trade-ins. The District completed the changeover two years ago. At the present time, all of the District's 43 vehicles are provided with two-way radios.

In considering the use of two-way radios, a district must keep in mind the public it serves. Today the mosquito abatement district must be alert to public opinion and the effects of that opinion on its program. Two-way radio is a valuable tool in molding that opinion by giving the public fast and efficient service.

## DESIGN AID FOR LAND GRADING

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The development of irrigation systems in California has been accompanied by land grading and related improvements to increase the area of irrigable land and to allow more efficient use of available water. Since the first land was graded or leveled for irrigation, numerous methods have been devised to assist farmers, engineers, and contractors in designing proper field grades and computing earthwork quantities. Design methods are generally tedious and time consuming, particularly when attempts are made to arrive at a field grade which is the most economical, i.e., one in which earthwork has been reduced to a minimum.

Increased emphasis on mosquito source reduction through physical changes has greatly increased the need to utilize advanced technology in solving engineering problems. It is within the capability of many local mosquito control agencies to provide technological assistance as an incentive to the growers for improving their property to achieve better production and, at the same time, mitigate existing mosquito problems. We anticipate that in future years more agencies will employ personnel capable of gathering field data and other necessary information for landleveling designs. This will facilitate the working relationship between local agencies and Bureau of Vector Control and Solid Waste Management staff in processing data required in the solution of problems.

Several factors which influence field and grades are:

1. Grower needs and future plans.
2. Topography.
3. Field Geometry.
4. Irrigation (type and quantity of water available).
5. Drainage (type, capacity, etc.).
6. Soil type, depth to hardpan, permeability, water table.
7. Crops.

These and other factors must be considered in developing alternative designs whether the work is done by hand or machine methods.

Collins and Shockley (1970) developed a computer program for plane surface design to facilitate land grading for irrigation. A major advantage of such a program is the time savings that can be realized from its use. The computer services section of the California Department of Public Health has taken this basic program and modified the language for use on the Department's RCA Spectra 70/45 computer in Berkeley. However, the capabilities of the program have not been changed and the input and output data are essentially the same.

The program has the capability of preparing several alternative designs for prescribed parameters within certain restrictive conditions. The program will compute cuts and

fills to establish a plane surface on any shaped field. Cuts and fills are balanced for minimum yardage movement within the limits prescribed for the job. The program computes the earthwork volumes and prints a grid sheet that can be used in marking cut and fill stakes for construction. The program also computes field area in acres. (Figures 1, 2, and 3.)

Input data can be given in the form of ground elevations on a grid pattern. Grid spacing must be at least 10 feet but not more than 500 feet. A maximum 52 grid points may be used on either of two axes. Odd shaped areas outside of the rectangular grid are called fringe areas, and can be described by the use of intermediate stations or grid points.

The series of events which must take place to utilize this computerized design aid are:

1. Obtain relative field elevations, soil depths, head of water and drainage requirements.
2. Determine parameters which must not be exceeded by the final elevations.
3. Prepare data for computer services. This consists of entering data on input sheets from which a keypunch operator can prepare the computer cards.
4. Evaluate the output to determine if the desired results were obtained or if changes are necessary.
5. Use the computer output for construction.

Mosquito abatement personnel can readily perform the first and last procedures of the foregoing events. These steps require many man-hours and thus represent considerably more money than keypunch and computer time.

An example of a typical regrading problem is presented. A planetable survey of the 32-acre field was made with the assistance of district personnel. Input data were prepared in the BVC/SWM office in Sacramento and several alternative solutions were requested. These alternatives were explored because the owner was uncertain about future use of the property. A typical run considering two design alternatives used a few seconds less than 8 minutes of computer time for a cost of \$16.69. Approximately 3 hours of keypunch and verification time were required at about \$10/hr, including overhead, bringing the total cost to approximately \$47. This was slightly less than \$1.50/acre. Once the field data cards are keypunched, other alternative designs can be requested and costs are usually less than \$1/acre. Cost per acre becomes less as field size increases.

Availability of this design aid through the Department's computer services can be considered a major asset to California's mosquito control agencies. Arrangements are possible whereby the keypunch and computer costs can be billed

to the districts. The BVC/SWM offices in Sacramento and Fresno are available for technical assistance in gathering data, organizing and submitting the data, interpretation of the results and using these results in the field.

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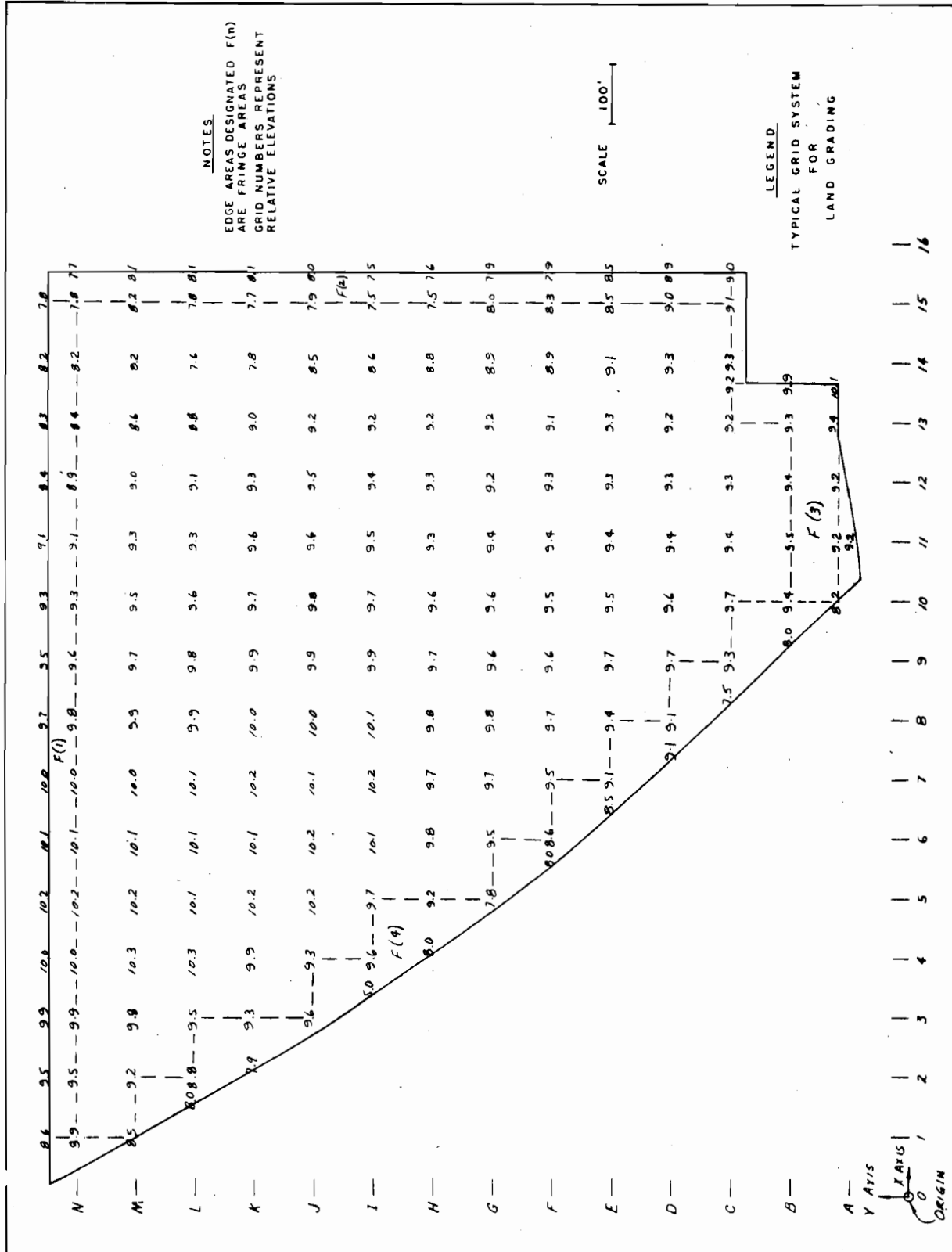


Fig. 1.—Typical grid system for land grading.

CUT VOL=14124.2 CY

FIELD AREA = 31.98 AC

AVERAGE EXCAVATION = 442. CY/AC

FILL VOL = 9560. CY

C/F RATIO = 1.48

MAX CUT = 1.04 FT

MAX FILL = 2.43 FT

## FRINGE POINTS NOT ON GRID COORDINATES.

X,Y COORDINATES	ELEV	GRADE	C/F
1+00 , 14+40	8.6	9.6	F 1.0
2+00 , 14+40	9.5	9.5	C 0.0
3+00 , 14+40	9.9	9.4	C 0.5
4+00 , 14+40	10.0	9.3	C 0.7
5+00 , 14+40	10.2	9.2	C 1.0
6+00 , 14+40	10.1	9.1	C 1.0
7+00 , 14+40	10.0	9.0	C 1.0
8+00 , 14+40	9.7	8.9	C 0.8
9+00 , 14+40	9.5	8.8	C 0.7
10+00 , 14+40	9.3	8.7	C 0.6
11+00 , 14+40	9.1	8.6	C 0.5
12+00 , 14+40	8.4	8.5	F 0.1
13+00 , 14+40	8.3	8.4	F 0.1
14+00 , 14+40	8.2	8.3	F 0.1
15+00 , 14+40	7.8	8.7	F 0.4
15+40 , 14+00	7.7	8.2	F 0.5
15+40 , 13+00	8.1	8.3	F 0.2
15+40 , 12+00	8.1	8.4	F 0.3
15+40 , 11+00	8.1	8.5	F 0.4
15+40 , 10+00	8.0	8.5	F 0.5
15+40 , 9+00	7.5	8.6	F 1.1
15+40 , 8+00	7.6	8.7	F 1.1
15+40 , 7+00	7.9	8.8	F 0.9
15+40 , 6+00	7.9	8.9	F 1.0
15+40 , 5+00	8.5	9.0	F 0.5
15+40 , 4+00	8.9	9.1	F 0.2
15+40 , 3+00	9.0	9.2	F 0.2
13+60 , 3+00	9.2	9.4	F 0.2
13+60 , 2+00	9.9	9.5	C 0.4
13+60 , 1+20	10.1	9.6	C 0.5
13+00 , 1+20	9.4	9.6	F 0.2
12+00 , 1+20	9.2	9.7	F 0.5
11+00 , 1+20	9.2	9.8	F 0.6
10+00 , 1+20	8.2	9.9	F 1.7
9+40 , 2+00	8.0	9.9	F 1.9
8+40 , 3+00	7.5	9.9	F 2.4
7+50 , 4+00	9.1	9.9	F 0.8
6+60 , 5+00	8.5	9.9	F 1.4
5+70 , 6+00	8.0	9.9	F 1.9
4+80 , 7+00	8.0	9.9	F 1.9
4+20 , 8+00	8.0	9.9	F 1.9
3+50 , 9+00	8.0	9.8	F 1.8
2+80 , 10+00	8.0	9.8	F 1.8
2+20 , 11+00	7.9	9.8	F 1.9
1+60 , 12+00	8.0	9.7	F 1.7

Fig. 2.—Fringe area computer printer.

PROJECT CSLA-72-20, E 1/2, S 35, T18N, R14W  
 TERREL FARMS - IRRIGATED PASTURE

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	
14	8.9 9.6 F 0.7	9.5 9.5 C 0.0	9.9 9.4 C 0.5	10.0 9.2 C 1.0	10.2 9.2 C 1.0	10.1 9.1 C 1.0	10.0 9.0 C 1.0	9.6 8.9 C 0.9	9.6 8.8 C 0.8	9.6 8.7 C 0.6	9.3 8.6 C 0.5	9.1 8.5 C 0.4	8.9 8.5 C 0.4	8.4 8.4 C 0.0	8.2 8.3 F 0.1	7.8 8.2 F 0.4
13	8.5 9.7 F 1.2	9.2 9.6 F 0.4	9.8 9.5 C 0.3	10.3 9.4 C 0.9	10.2 9.3 C 0.9	10.1 9.2 C 0.9	10.0 9.1 C 0.9	9.9 9.0 C 0.9	9.7 8.9 C 0.8	9.5 8.8 C 0.7	9.3 8.7 C 0.6	9.0 8.6 C 0.4	8.6 8.5 C 0.1	8.2 8.4 F 0.2	8.2 8.3 F 0.1	9.2 8.3 F 0.1
12	8.8 8.9 F 0.9	9.5 9.6 F 0.1	10.3 9.8 C 0.5	10.1 9.3 C 0.8	10.1 9.3 C 0.8	10.1 9.3 C 0.8	10.1 9.2 C 0.8	9.9 9.1 C 0.8	9.8 9.0 C 0.8	9.6 8.9 C 0.7	9.3 8.7 C 0.5	9.1 8.5 C 0.4	8.8 8.5 C 0.2	8.8 8.5 F 0.1	7.6 8.5 F 0.6	7.8 8.4 F 0.6
11	9.3 9.7 F 0.4	9.9 9.6 C 0.3	10.2 9.5 C 0.7	10.1 9.4 C 0.7	10.2 9.5 C 0.7	10.1 9.4 C 0.7	10.2 9.3 C 0.9	10.0 9.2 C 0.8	9.9 9.1 C 0.8	9.7 9.0 C 0.7	9.6 8.9 C 0.7	9.3 8.8 C 0.5	9.0 8.7 C 0.3	8.6 8.6 F 0.8	7.7 8.5 F 0.8	7.9 8.6 F 0.7
10	9.6 9.8 F 0.2	9.3 9.7 F 0.4	10.2 9.6 C 0.6	10.2 9.5 C 0.7	10.2 9.5 C 0.7	10.2 9.5 C 0.7	10.1 9.4 C 0.7	10.0 9.3 C 0.7	9.9 9.2 C 0.7	9.8 9.1 C 0.7	9.6 9.0 C 0.6	9.5 8.9 C 0.6	9.2 8.8 C 0.4	8.5 8.7 F 0.2	8.5 8.6 F 0.7	7.9 8.6 F 0.7
9	9.6 9.8 F 0.2	9.7 9.6 C 0.0	10.1 9.6 C 0.5	10.1 9.6 C 0.5	10.1 9.6 C 0.5	10.1 9.6 C 0.5	10.2 9.5 C 0.7	10.1 9.4 C 0.7	9.9 9.3 C 0.6	9.7 9.2 C 0.5	9.5 9.0 C 0.4	9.4 8.9 C 0.4	9.2 8.9 C 0.3	8.6 8.8 F 0.2	8.6 8.7 F 1.2	7.5 8.6 F 1.2
8	9.2 9.6 F 0.8	9.8 9.7 C 0.1	9.7 9.6 C 0.1	9.8 9.7 C 0.1	9.8 9.7 C 0.1	9.8 9.7 C 0.1	9.7 9.6 C 0.3	9.6 9.5 C 0.3	9.7 9.4 C 0.3	9.6 9.3 C 0.3	9.3 9.2 C 0.1	9.3 9.1 C 0.2	9.2 9.0 C 0.2	8.8 8.9 F 0.1	8.5 8.8 F 1.3	7.5 8.8 F 1.3
7	7.6 9.9 F 2.1	9.5 9.8 F 0.3	9.7 9.6 C 0.0	9.7 9.6 C 0.2	9.7 9.6 C 0.2	9.7 9.6 C 0.2	9.6 9.5 C 0.1	9.6 9.5 C 0.1	9.6 9.5 C 0.1	9.6 9.4 C 0.2	9.4 9.3 C 0.1	9.4 9.2 C 0.1	9.2 9.1 C 0.1	8.9 9.0 F 0.1	8.9 9.0 F 0.9	8.0 8.9 F 0.9
6	8.6 9.9 F 1.3	9.5 9.8 F 0.3	9.7 9.6 C 0.0	9.7 9.6 C 0.0	9.7 9.6 C 0.0	9.7 9.6 C 0.0	9.6 9.5 C 0.0	9.6 9.5 C 0.0	9.6 9.5 C 0.0	9.5 9.4 C 0.0	9.4 9.3 C 0.0	9.4 9.2 C 0.0	9.3 9.2 F 0.1	8.9 9.1 F 0.2	8.9 9.0 F 0.7	8.3 9.0 F 0.7
5	9.1 9.9 F 0.8	9.7 9.6 F 0.1	9.5 9.4 F 0.1	9.5 9.4 F 0.1	9.5 9.4 F 0.1	9.5 9.4 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.1 9.2 F 0.1	8.5 9.1 F 0.6	8.5 9.1 F 0.6
4	9.1 9.8 F 0.8	9.6 9.5 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.2 9.1 F 0.1	9.2 9.1 F 0.1	9.0 9.1 F 0.1	9.2 9.2 F 0.2	9.0 9.2 F 0.2
3	9.1 9.8 F 0.8	9.6 9.5 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.2 9.1 F 0.1	9.2 9.1 F 0.1	9.0 9.1 F 0.1	9.2 9.2 F 0.2	9.1 9.3 F 0.2
2	9.1 9.8 F 0.8	9.6 9.5 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.4 9.3 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.3 9.2 F 0.1	9.2 9.1 F 0.1	9.2 9.1 F 0.1	9.0 9.1 F 0.1	9.2 9.2 F 0.2	9.1 9.3 F 0.2

Fig. 3.—Grid system computer printout.



## SUCCESSFUL SOURCE REDUCTION ON TIDAL SALT MARSHES

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### ABSTRACT

In 1968, Marin County Mosquito Abatement District initiated changes in its methods in order to achieve better mosquito control with less pesticide. Labor deployment, pest annoyance thresholds, surveillance and source reduction were improved and heavily stressed. Within four years several major mosquito problems were eliminated and pesticide use was reduced by over 80%. In 1972, only 9 gallons of fenthion were used for all mosquito suppression.

The single activity contributing most importantly to this achievement was the District's source reduction program on

over 5,000 acres of tidal salt marshes. With the use of a Spryte all-terrain vehicle and a Speedscavel ditcher (Thiokol Corporation, \$15,000) mosquito breeding sources on the marshes were connected to sloughs that empty into the bay. Flooding tides no longer impounded and *Aedes dorsalis* mosquitoes were reduced to numbers which would not invade neighboring communities. Costs for *A. dorsalis* control dropped from \$4,735 in 1968 to \$7 in 1971. Taking into account maintenance costs, the Spryte and Speedscavel will pay for themselves in less than five years.

## MOSQUITO SOURCE REDUCTION HELPS CUT DISTRICT TAX RATE

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Consolidated Mosquito Abatement District has enjoyed the experience of an almost continuous declining tax rate from the beginning of its operations. The annual tax rate that started off at 15 cents is down to only 7½ cents this fiscal year. Cutting the tax rate in half over a 27 year period is rather slow progress, but it is progress. To add to the nice feeling, preliminary estimates indicate the District's tax rate will be cut again next year. Hopefully the decline will not continue at the same rate during the next 27 years. A zero rate would not allow very high wages.

One question that will be raised when a tax supported agency lowers its rate is, was it too high in the beginning for the services required? This can be answered at least in part by the events that occurred at Consolidated MAD.

The District formed in June 1946 by the Fresno County Board of Supervisors after petition by interested citizens started operation on borrowed money. Formed too late to be on the tax rolls for fiscal year 1946-47, the Board of Trustees had no hesitation over going to the bank for a loan to get the work started. They had the security of a firm commitment from the Board of Supervisors that a double rate could be levied in fiscal year 1947-48. The rate for that year was 30 cents. This made it possible to pay off the bank loan, meet other expenses of getting started and end up with a nice cash carry-over. In fact the reserves were so large it was decided to lower the tax rate for 1949-50 to 12 cents. It was soon evident this rate would not bring in enough to meet current needs and maintain adequate funds to start the new fiscal year. The rate was raised to 15 cents for fiscal year 1951-52 and remained there until fiscal year 1955-56 when the decline started from a more established position.

While the early false start in reducing the tax rate did indicate the financial need for that time was greater than had been estimated, it also revealed the attitude of the trustees on the responsibility of the District. This was to provide the most efficient service practical to the taxpayer. It was accepted from the beginning that this could be done best by reducing known mosquito sources to the smallest size feasible. A policy was adopted at the August 1947 Board meeting fixing responsibility for correcting mosquito sources on the land owner. Sources that could reasonably be considered natural would be the responsibility of the District. The pursuit of this policy has been constant and the results have been an important factor in the declining tax rate.

Another question that could be raised on the effort to keep bringing the tax rate down is, were employee salaries and benefits sacrificed? While these are never high enough to meet the needs of the average employee they were in line with similar jobs in the area. In the recent, "California Mosquito Control Association Salary & Working Conditions Schedule effective August 1, 1972", Consolidated shows quite well in all categories for the south San Joaquin Valley.

Salaries will never match the Bay Area or Southern California but those workers don't have the advantages of living in Fresno County.

A question might be raised on the quality of the District's physical assets but, here again they compare favorably with similar agencies in the area. As the name implies Consolidated is made up of many parts. There were four incorporated cities in the original formation and the safe course was to not identify the agency title with any one community. This had even more credence as annexations added four more cities. This compromise on the name did not, however, carry over to the desire by each trustee to have local service. Selma was accepted as the location for the headquarters office mainly because the first manager (Ed Davis) lived there. As the big annexations of the Reedley High School District and the Clovis High School District were approved and full-scale field operations started in 1947, depots were established in each of the cities within the District. Some of these were combined in later years but there are still six depots that are constantly being modernized. Equipment is standard, ranging from hand-guns to aircraft with the one man in a jeep the standard field inspection-treatment unit. INSPECTION, to assure proper timing for treatment, is the WORD at Consolidated MAD.

The decline in the tax rate has not meant a decline in the effectiveness of the control program. While adult mosquito populations have been a little higher in the last two seasons more money would not have changed the trend. Spray materials just were not working as well as they had in the past and adjustments were not possible overnight. The main adjustment will be requiring more source reduction so for the long range, having fewer chemicals to work with will be a benefit.

Like the mosquito population, failure of the chemical control program has confused the figures on source reduction. The bottom just about dropped out of the "acres sprayed" graph in 1972. Total area sprayed last year was down to an all time low of 14,770 acres. Credit can be taken for source reduction accounting for part of the big drop from 41,710 acres in 1971 but better timing of sprays and just not spraying under certain conditions also contributed to the decline.

Realistically, the steady improvement, modifications, elimination or whatever of mosquito sources through the years has been the main factor in the District's progress. A direct attack has been made on all types of mosquito sources. While sources related to irrigation for agriculture are still number one in number and severity, the greatest amount of area eliminated has been in this category. Industrial liquid waste has not posed a problem for years and community sewer disposal sites are getting close to the almost zero problem. Household and street problems still take a lot of time but this is more in looking, rather than

treating. Natural sources have been corrected and maintained so that fish and other predators keep mosquito larvae populations down to a reasonable level. Fish are also a must in all man made water holding structures. Even if there is a suspicion that water will remain for any length of time fish are planted in late winter or early spring. This may not

be true source reduction but it does reduce the need to spray and cuts the bill for insecticides. This, of course, reduces the budget and this reduces the tax rate. Cutting the District rate to one-half the present rate would be a nice goal to shoot for.

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# NEW MOSQUITO SOURCES AND THEIR PREVENTION IN URBAN AREAS

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I would like to introduce this subject by quoting the following paragraph which best describes the factors that have brought our District to this point:

"In brief, our mosquito population has greatly decreased, but the number and diversity of sources has increased tremendously. As a result, we are confronted with a wider range of species, requiring sharper control precision and efficiency. Fortunately, along with an increasing population comes an increase in assessed valuation, making funds available for better materials and equipment, and for the development of improved techniques. In other words, with urbanization one can expect new problems as well as the means for answering those problems." (Mathis 1962)

In an effort to improve the urban and suburban environment and to become environmentally aware and sensitive, public and private agencies are creating new aquatic habitats that are ideal for breeding mosquitoes and other aquatic nuisances. The creation of these aquatic habitats is also due to a demand by the public for more aquatic sources for recreational, environmental and aesthetic uses. These agencies are in fact creating more aquatic habitats than this environment has had in the last hundreds of years. These new aquatic sources are quite varied being either large or small and either functional or aesthetic. The mosquito and other aquatic nuisance problems are as varied as these sources and the problems emanating from just one type of these sources, man made recreational lakes, has been documented by Magy (1968).

DESCRIPTION OF SOURCES.—I have arbitrarily distinguished these sources in which my District has come in contact into three categories.

## 1. Ornamental Aquatic Habitats

A. Living Complexes: The ornamental aquatic habitats within living complexes such as apartments and condominiums are quite varied consisting of either a large single pond, several small ponds, ponds connected by streams and waterfalls or any combination thereof. They are constructed of cement, gunite or soil cement and formed to any shape. These sources can look like natural ponds and streams or reflecting pools and can be used for only beautification or many purposes such as fishing. At present we have 30 of these sources mapped throughout Orange County and are finding more under construction every month.

B. Recreational or Real Estate Lakes: These lakes and their problems have been well documented with respect to their biological problems by Magy (1968) and their planning and management problems by Rickert (1971). These lakes are usually at least five acres in size with the maximum amount of shoreline being created by the design of these lakes. Presently we have five of these lakes in operation and two proposed for construction in Orange County.

C. Open Space Demands: The development of areas for open space, parks and recreational potential can significantly increase the size and number of sources. This includes enhancement of natural water courses and the use of excess lands along flood zones and river levees. These would be large greenbelt areas which would include many aquatic habitats. At present there are three plans being implemented in Orange County, the Santa Ana-Santiago Creek Greenbelt Plan, the Aliso Creek Greenbelt Plan and the Huntington Beach Central Park Project.

## 2. Environmental Education Centers

These centers are being designed and built on public school properties for the outdoor environmental education and some are being financed with federal funds. At present there are four of these centers in operation and several more under construction. Each one of these centers includes aquatic habitats as a major theme in their design. These can be designed and operated without a mosquito nuisance being created. Also, they can be an excellent example for public education in mosquito control.

## 3. Subsurface Transformer Enclosures

Public utility companies in their cooperative programs with local governments to make a more pleasing visual environment, are putting residential utility services underground. This involves electric lines and transformers which are encased in a concrete housing for protection. The transformers are set in cylindrical concrete vaults seven feet deep, the tops of which are usually at ground level with only a metal grate covering the openings to provide air ventilation for cooling. The vaults are usually located near the sidewalk and serve four houses.

In December 1970 the District discovered several of these enclosures breeding mosquitoes. In 1971 Madera MAD discovered a number of the enclosures breeding mosquitoes and contacted the Bureau of Vector Control and Solid Waste Management for assistance. At present, we have mapped 988 of these enclosures within the District. Myers, Colson and Challet (1973) have adequately described these sources and the steps that have led to the preventive action.

We anticipate expansion of sources such as these and some with which we have not come in contact. This is underlined by the fact that Southern California will receive operational use of California Water Project water in 1973. Also, many of our inland waste water plants are using recycled water for the same purposes as mentioned above.

PREVENTIVE ACTION.—Preventive action takes place while the source is still in the planning stage. This requires a knowledge of what is being built in your district and this

is by whatever means you might use. This knowledge can come from your contacts with other governmental agencies such as land and water use agencies, planning, building or environmental departments, newspapers or surveillance. The district would like to advise the builders and developers of potential mosquito breeding sources. This means, "changing the method of the administration of an abatement program from working with the property owner on an operational basis to a preventive program of working with planning and development agencies." Ideally, agencies concerned with development, such as planning commissions and city councils should have the developer contact the District for guidelines for keeping these potential sources free of mosquito breeding. This does not mean checking plans for each project, but providing printed guidelines or specifications for design and maintenance.

Fowler (1962) states, "... three times as many control operators are required in areas that have become urbanized," and this has been the experience of the Orange County MAD. With this statement in mind, can the addition of a new staff member or the redirection of a present staff member to monitor these new sources in their planning stages be economically feasible? Would this new function reduce future manpower needs for a district with an expanding urban area? Grant (1966) stated, "For every preventive measure planned and executed in urban water management, we have a host of problems created through the economy." This is true in a number of cases in this District but a preventive program should be tried.

**Living Complexes:** When our operators first found that these sources were causing nuisances, we monitored one of these sources from which we had the most service requests. This was an older complex which had poor maintenance procedures. The mosquito and chironomid midge problems reached a point where the owner did not want to continue control or maintenance so the source was filled with sand.

Two cities have had the developers of proposed projects contact the District about the prevention of mosquito breeding. At the request of the architect of the first development we provided a set of "Proposed Specifications for the Prevention of a Mosquito Nuisance in Proposed Aquatic Habitats." These specifications can be sent to architects, planners and building inspectors.

**Specifications** – We hope to submit these specifications to the planning and building departments of the various cities and the County, so that they may in turn provide these to those persons that might include aquatic habitats on their property.

The problems of chironomid midge nuisance in these sources is still a problem and only proper maintenance and abatement action seem to be the answer at this time. Some guidelines have been arrived at through the little experience that we have had:

## Orange County Mosquito Abatement District PROPOSED SPECIFICATIONS FOR PREVENTION OF MOSQUITO NUISANCE IN PROPOSED AQUATIC HABITATS

Mosquitoes can be produced in any situation where standing water occurs.

The best way to prevent mosquito production in any water source is to design, operate and maintain that water source so as not to produce mosquitoes. It is the property owner's responsibility to see that a public nuisance from mosquitoes is not created on his property according to the California Health and Safety Code, Section 2274. It is the property owner's responsibility to abate the nuisance as stated in Section 2275.

Instituting the following measures in each water source should provide a mosquito-free environment.

1. Provide moving water – this creates a flow which prevents larval production and egg laying.
2. Prevent emergent vegetation – prevention of emergent vegetation or overhanging vegetation will reduce the amount of habitat for the mosquito larvae in the water.
3. Prevent runoff – runoff from banks into ponds and streams increases available food for the mosquitoes.
4. Introduction of mosquito fish – if applicable to the source.
5. Proper maintenance – each facility must be maintained so that mosquito production does not occur.
6. Larviciding when required – when and if mosquito production occurs, then the appropriate larvicide shall be used.

Each source is different and unique so that mosquito control measures have to be suited to that individual source.

The Orange County Mosquito Abatement District will provide consultation and advice on the design, operation and maintenance of these habitats.

**Recreational and Real Estate Lakes:** Our experience with these small lakes has been limited in time and to operational abatement actions. However, the newer lakes have had this experience to use for design and operation so that some newer lakes have had less of a problem with both mosquitoes and midges. Specifications by Magy are adequate but only reduce the problem somewhat. Mosquito breeding is reduced because of algae control and the introduction of fish. However, chironomid midge nuisance is very great until the lake gets stabilized.

**Open Space Demands:** These parks and greenbelt areas are just getting started and we have no idea as to what will be built in the aquatic habitat areas. They will probably be similar to golf course water hazards. These sources will have both mosquito and midge problems. These sources can, however, be designed and maintained so no mosquito problems occur. Specifications should be included in a mandatory maintenance agreement when the building permit is issued. We have been in contact with the coordinators of these plans and are included on their contact lists.

**Environmental Education Centers:** Our experience with these sources has been quite limited. We have contacted each school about this problem and also the Orange County Education Department which acts as advisor. These can also be designed so that little or no mosquito breeding will occur. These centers can be of value as an educational source for mosquito biology and control very close to the students. The Orange County Education Department has offered to help us in this respect.

**Subsurface Transformer Enclosures:** With the cooperative help of the Bureau of Vector Control and Solid Waste Management on a statewide basis and our local utility companies, a statewide program has been started on preventive action before more are built. Abatement of these sources will take place on an individual source basis.

Control operators are finding these new sources continuously and a great many of them have had nuisance problems. We should try to prevent these from being nuisance liabilities but rather an asset to the community.

First, we must find out when and where these sources are going to be built. Secondly, we must offer guidelines to the developers on how these sources should be built to prevent aquatic nuisance problems. This can be done by submitting these guidelines to the agencies with responsibility over land use permits. We should then help to see that potential mos-

quito breeding sources are built so as not to produce mosquitoes. This can be done by including your guidelines on a project in the Environmental Impact Statement. These statements can be used to a district's advantage providing you have the data to support your stand. Each governmental jurisdiction by law must have a general plan for land use and included within these are open space and conservation elements. These are areas where input from a mosquito abatement district could offer preventive mosquito control on proposed aquatic sources. There is a possibility that coastal mosquito problems might be helped by being put on a preventive basis with the appointment of the Coastal Conservation Commissions.

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## TOLERANCE THRESHOLDS IN MOSQUITO PEST MANAGEMENT

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An entomological and sociological study is being conducted to study the extent of discomfort inflicted by pest mosquitoes. The functional dependence of urban and rural residents' response to weekly population indices of five different mosquitoes has been studied continuously in Sutter County, California, since 1970. Measuring public response to pest mosquitoes by complaints or service requests registered to the local mosquito abatement district

office did not appear to differ significantly from results of personal interviews of randomly selected urban and rural residents by student survey workers. Results of preliminary analysis indicated that *Anopheles freeborni* accounted for the major response of urban residents; *Culex* and *Aedes* species, for rural residents. The extent to which seasonal changes in the mosquitoes' age, physiological state and feeding habit affect their ability as human pest is still being investigated.

## PAROUS RATES OF *ANOPIHELES FREEBORNI* POPULATIONS IN NORTHERN CALIFORNIA

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**INTRODUCTION.**—The physiological aging of mosquito populations is used to assess vector potential, to evaluate control efficiency and to provide population data for life tables and computer simulation models. A number of aging techniques have been developed and applied to mosquitoes. However, only with Polovodova's technique (Detinova 1962) can parous females be separated into distinct age classes. In this technique, the individual ovarioles within ovaries are examined for dilatations (follicular scars). Each dilatation marks the completion of one gonotrophic cycle by the female. If oogenesis is synchronous and if the duration of the gonotrophic cycle is estimated, the chronological age can then be approximated from the physiological age.

For the past several years now we have been using Polovodova's technique as a tool for detecting seasonal changes in the age structure of *Anopheles freeborni* in the Sacramento Valley. In 1969, we found that the technique was applicable to *A. freeborni* populations and the results of the preliminary dissections of field populations were presented at these meetings in 1970 (McKenna and Washino 1970). Later in that same year, the technique was used as an aid for evaluating the effectiveness of ultra-low volume spraying for control of *A. freeborni* in Colusa County (Washino et al. 1972).

Our earlier investigations were severely limited because females were collected from only a few locales and because only small samples of the collected females were dissected. In order to rectify these limitations, an expanded study was initiated during the summer of 1972 in which mosquitoes from 23 sampling sites were processed by three dissectors. The two dissectors who had not worked on the earlier study spent three weeks dissecting females of known age and then passed (at better than 95% efficiency) a test of unknown nulliparous and parous individuals prepared by one of the authors.

**MATERIALS AND METHODS.**—Adult mosquitoes were collected from 24 red boxes placed at the 23 localities in Sutter and Colusa counties. Approximately half the red boxes were collected on Tuesday mornings and the other half on Thursday mornings for an 11-week period from the first week of July until mid-September.

On all occasions the mosquitoes were classified and dissected during the afternoon of the day of collection. Mosquitoes were classified according to species, to sex, and in the case of females, to physiological state (empty, blooded or gravid). The ovaries of females were dissected and the Christopher's stage of the oocytes and the level of parity were recorded. Except on three occasions when the number of females was prohibitive, the entire collection of female mosquitoes from red boxes were processed. On those three occasions, approximately 70 to 88 percent of the total sample was aged.

Data from the classifications and dissections were transferred to computer punch cards and analyzed by a multiple cross tabulation program based on sampling technique, locale, date, species, sex, and physiological state.

**RESULTS AND DISCUSSION.**—A total of 6931 *A. freeborni* females was collected from the red box stations during the study period. We were able to dissect and age 6189 of these females. Table 1 shows the results of the dissections on a monthly basis.

Table 1.—Monthly age composition of *Anopheles freeborni* in the Sacramento Valley, Summer of 1972.

Month	No. dissected	% NP	% 1P	% 2P	% 3P	% 4P	% 5P
July	2533	70.8	27.7	1.3	.1	0	0
August	2549	68.6	27.4	3.4	.5	0	0
September	1107	78.5	17.5	3.4	.2	.2	.1

The highest overall parity was recorded in mid-August although the oldest individuals (4 and 5 parous) encountered were in late August and early September.

A marked increase was observed in empty females in late August and in the beginning of September. This increase was attributed to the emergence of the overwintering females because during this same period the proportion of these empty females which were nulliparous with oocytes in Stage I-II studies have shown that the diapausing females of *A. freeborni* are empty nulliparous with the resting stage of the ovarioles in I-II rather than the normal Stage II of gonotrophic females (McKenna and Washino 1970).

In summary, our data from the physiological aging of over 6,000 *A. freeborni* females during the summer of 1972 suggest that the oldest and consequently most dangerous females in terms of vector potential occur in late August and early September. During this same period, however, because of the emergence of the overwintering population, these potentially dangerous females represent but a small proportion of the total population. Much older females than had previously been reported for this species do exist, at least in September populations.

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# RESULTS OF PRELIMINARY MARK – RELEASE – RECAPTURE TRIALS WITH *Aedes nigromaculis*

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Increasing concern with biological, ecological and genetic control methods, which are highly density dependent, creates a greater need for knowledge of absolute population parameters such as survival, fecundity and dispersal. These are required firstly to estimate the ratio of agents needed to be released relative to the size of the population of the target organism, and secondly to monitor population levels during or subsequent to the release, as well as dispersal into and out of the target area.

One well known method is of course mark-release-recapture. To provide statistically reliable data multiple releases must be made over consecutive days or other regular intervals. Each separate release must thus be marked with a distinct code. In order to get information on dispersal there must be more than one distinct release and or catch site. Ideally a ring of catch and release points could be used so that information could be gained on directional trends of dispersion. Assuming a minimum of say 5 sites over a 10-day period we clearly need 50 different marks.

Large animals and even large insects offer fewer problems for multiple marking. Gary (1971) has, for example, glued numbered metal disks to bees and such a method might be extended to large biting flies such as tabanids or *Glossina*. Working with small populations of the yellow fever mosquito, Sheppard et al. (1969) in Thailand and McClelland and Conway (1970) in Tanzania were able to hand-paint number codes on individual mosquitoes, because single-day catches were less than 50. In these cases high recapture rates provided adequate data from the small samples.

The dense and extensive populations of many mosquitoes, particularly those of ricefields and irrigated pastures such as *Aedes nigromaculis*, preclude individual handling. Marks need to be applied quickly to huge numbers of insects *en masse* in order to yield a useful number of recaptures. Known marking substances, however applied, fall into three groups: radioactive trace elements, dyes and fluorescent dusts (Southwood 1966). All three have the serious limitation that the number of clearly distinguishable marks is, most optimistically, below ten. Radioisotopes suitable for marking live insects have the disadvantage of short half-life. Neither dyes nor isotopes can be identified without special treatment. An entire catch must be processed with solvent or taped to X-ray film in order to recognize a possibly minute proportion of recaptures. Fluorescent dusts have been widely used and have the great advantage, over radioisotopes and dyes, in that marked insects can be recognized

alive by brief exposure to long wave ultra violet "black light". Unmarked mosquitoes in the catch can thus be marked and released to increase the efficiency of the operation. Again the number of dusts that fluoresce well and are distinguishable is less than ten.

**METHODS AND DISCUSSION.**—Rationale — The analytical method used in the present study owes its development to the improvement of techniques for detecting trace contaminants as indicators of atmospheric pollution. Thus you can say it is a byproduct of the ecology movement.

Minute quantities of heavier elements can be detected and identified by their characteristic X-ray emission spectra when bombarded by alpha particles or an electron beam. Suitable trace-element compounds can thus be added to a fluorescent marking dust in various code combinations. Such elements must 1) be not detected in unmarked insects, 2) be insoluble, stable and unreactive, 3) produce identifiable X-ray spectra in the presence of X-ray emission from other marker elements, natural elements present in the specimen and that from the marker dust. Where zinc sulfide is used as the visual marker its presence is confirmed by its X-ray spectrum. Naturally occurring fluorescence such as that reported by Reeves et al. (1948) thus can cause no problem. Further details of the method have been described by McClelland, McKenna and Cahill (1973).

Mosquitoes were dusted with a mixture of the zinc sulfide powder found to be brightest by Bailey et al. (1962), Helecon® no. 1953, and combinations of three of the following non-radioactive trace elements: zirconium oxide, uranium oxide, molybdenum (elemental), niobium oxide, lanthanum carbonate and cerium oxide.

For decoding, marked mosquitoes were killed and fixed to thin Mylar® film. Protected in this way they can be kept indefinitely since the atoms are completely stable. Field work and code analysis can thus be conveniently separated in space and time yet the progress of an experiment can be immediately known from the rate of recapture. Although the trace elements and the method of analysis are relatively costly and require the facilities of a cyclotron, it should be remembered that only known recaptures are tested, so no time or money need be wasted on unmarked insects, and the samples can easily be shipped to a cooperating cyclotron facility.

When the samples are ready for testing at the cyclotron they are mounted on a rotatable target wheel or other sample-changing device, inside a vacuum chamber in the path of a focused beam of alpha particles generated by the cyclotron. All of the atoms in the path of the beam emit X-rays of certain energies characteristic of each element. Counts of the X-rays of each energy level are collected and analyzed by an on-line computer.

Field Trial — In July and August 1972, with the helpful cooperation of the Sutter-Yuba Mosquito Abatement Dis-

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Number caught per day	Number marked per day											Day		
935	820	-											1	
4042	816	19	19										2	
5308	3735	4*	4*	52	TOTAL RECAPTURED EACH DAY							3		
5028	2194	28*	2*	24*	78							4		
6732	2389					112						5		
4524	2084						66					6		
3057	1685							42				7		
2185	1374								28			8		
1731	981									47		9		
3058	2251										28	10		
4061	2651											20	11	
5855	3690												48	12
9412	--												180	13
55928	24670											735		

Fig. 1.—*Aedes nigromaculis*: partial results of a mark-release-recapture study, Sutter-Yuba County, California, July-August 1972.

\*numbers partially determined by x-ray analysis.

tract, we initiated a limited field trial to assess the potential of this technique under operational conditions. A site south of Marysville, California, was chosen where populations of *A. nigromaculis* were dense and subjected to little control.

A team of three catchers using battery-powered aspirators caught mosquitoes from their legs for a period of 75 minutes. The catch was divided into groups of about 25 mosquitoes and each was screened under black light. Samples containing marked individuals were taken back to Davis for separation of the recaptured individuals and analysis. The remainder was dusted with a differently coded fluorescent powder and released. The procedure was repeated for 13 days (Fig. 1). The mosquitoes released on day 12 were marked with Helecon no. 1767, a red-fluorescing dust. Thus it was known without X-ray analysis that 15 of the 195 mosquitoes recaptured on day 13 were more than 1 day old.

As can be seen from Figure 1 the recapture rate was good. The results of the cyclotron analysis were, however, disappointing but not discouraging. Much more dust was lost under field conditions than in the preliminary laboratory trials, as might have been expected. The quantities of trace elements were thus reduced below the threshold for detection. This limit is about 100 millionth part of a gram. Since we had added only about one part of the trace element to 50 parts of the fluorescent dust, the amount of trace element marker could be increased easily. We also ran into some unexpected problems. The clear plastic Krylon® fixative spray, used to mount the mosquitoes on the plastic film for analysis, was contaminated with lead, apparently from the solder in the aerosol can. The strong X-rays from the lead masked some of the weaker emission from the trace elements.

We are confident that the method can be refined to give the data needed. Nevertheless we are currently exploring the alternative of using coded mixtures of different fluorescent powders. The gross fluorescence would allow recogni-

tion in the field while identification of the code would be accomplished using an incident-light fluorescence microscope to recognize individual particles of the different powders.

#### Acknowledgments

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# SUSCEPTIBILITY OF ORGANOPHOSPHORUS RESISTANT *CULEX TARSALIS* TO EXPERIMENTAL INFECTION WITH WESTERN EQUINE ENCEPHALOMYELITIS VIRUS<sup>1</sup>

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## ABSTRACT

A study was conducted to determine if organophosphorus (OP) insecticide resistant *Culex tarsalis* was an efficient vector of western equine encephalomyelitis (WEE) virus. This study was prompted by the apparent correlation between the emergence of a highly resistant population of *C. tarsalis* to OP insecticides and the concomitant decrease in WEE viral activity in the Central Valley of California

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during 1969 and 1970. Susceptibility of adult female *C. tarsalis* to infection with WEE virus was determined by intrathoracic inoculation and by feeding on gauze pledgets soaked with a mixture of virus, defibrinated rabbit blood and sucrose.

No correlation was found between susceptibility to infection with WEE virus and resistance to five OP insecticides in *C. tarsalis* obtained from both field and laboratory sources. However, when colonized strains of OP resistant *C. tarsalis* and *Culex quinquefasciatus* were reared in the presence of a concentration of fenthion that initially killed 20 percent of the larvae, adult females of both species were more resistant to infection with WEE virus by intrathoracic inoculation than were nonpressured mosquitoes. The significance of these observations to encephalitis in California was discussed.

# OBSERVATIONS ON THE HATCHING OF *Aedes nigromaculis* (LUDLOW) EGGS: SEASONAL VARIATION

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The females of the pasture mosquito, *Aedes nigromaculis* (Ludlow), prefer to lay their eggs in bunch grass clumps in irrigated pastures, especially the clumps in depressions where moisture content is high and can be retained for a long period (Miura and Takahashi 1973).

Based upon laboratory hatching experiments and field observations, Telford (1963) reported that the hibernation of the pasture mosquito constitutes a true facultative embryonic diapause. He postulated that descending temperatures in the fall initiate diapause, whereas ascending temperatures terminate diapause in the spring. McHaffey (1972), also based upon laboratory tests, reported that eggs of the pasture mosquito entered diapause in response to a sub-optimum temperature (10°C, 50°F); the proportion of diapause was correlated to the length of exposure to the sub-optimum temperature. Maternal influence on egg diapause was also reported by Miura et al. (1968) and McHaffey (1972).

The purpose of this report is to present the results of hatching variation of *A. nigromaculis* eggs in different seasons. The results of a preliminary study on termination of diapause are also reported.

**MATERIALS AND METHODS.**—During summer and early fall, bunch grass clumps were collected periodically from irrigated pastures to observe influence of environmental conditions on egg hatchability. The clumps collected in early fall were also stored outdoors at the laboratory for further observations of influence of winter and spring conditions.

The general methods for determining hatchability were essentially the same as previously described (Miura and Takahashi 1973) with one exception that the samples were flooded 5 to 15 times at weekly intervals; each sample (a core 7 cm in diameter, or a piece of clump, ca. [ca.=circa=about] 50 cm<sup>2</sup>) examined was flooded in a white enamel pan (30 x 18 x 5 cm) for 48 to 72 hours at room temperature of 71.6° to 78.8° F (22° - 26° C) and RH 75 to 85%. The larvae were then counted by gently pouring the water into a clean white enamel pan. Newly collected samples from the field or outdoors were conditioned at least for 3 to 7 days at room temperature before being used for hatching tests. At the end of hatching tests, samples were then checked for unhatched eggs.

In order to study photoperiodic and thermoperiodic influence on termination of diapause two samples (ca. 50 cm<sup>2</sup> each) were collected from a bunch grass clump on January 12. One sample was wrapped with aluminum foil and the other was left unwrapped. The samples were then placed in a glass humidity chamber and then kept in a dual program (illumination, temperature) incubator for 49 days to reactivate the diapaused eggs. The incubator was pro-

grammed for daily cycle of 14 hr light at 80°F (16.7°C) followed by 10 hr darkness at 60°F (15.6°C). The original bunch grass clump used for this test was collected from the Costa Pasture, Hanford, Kings County on September 8, 1971 and has been stored outdoors at the laboratory.

**RESULTS AND DISCUSSION.**—During summer months (June, July, August, and September) over 90% of the eggs hatched at the 1st flooding; however, to obtain 100% hatch, at least 3 to 4 separate floodings were necessary (Table 1). This phenomenon of "delayed hatch" has been reported in the previous report (Miura et al. 1968) and it has been speculated that this mechanism may play an important role in the successful development of *A. nigromaculis* in temporary irrigation water.

Figure 1 shows the results of hatching tests of the pasture mosquito eggs collected from the field in different months together with data showing the average minimum and minimum temperatures and daily photoperiod. In summer months, temperature fluctuated between 85° to 55°F (29.4° - 12.8°C) and average daily sunshine was 12 hrs or more; under this condition the pasture mosquito eggs will hatch readily at the 1st flooding and the species will breed continuously if sufficient water is available.

The results of hatching tests during November, December, and January are shown in Table 2. At the 1st flooding very few eggs hatched (2 hatched out of 8,891 eggs); ca. 35% at the 2nd; 19% at the 3rd; 30% at the 4th flooding; and the remaining viable eggs hatched subsequent floodings. During this period, maximum and minimum temperature fluctuated between 65° to 32°F (18.3° - 9°C); daily photoperiod varied 8 to 4.5 hrs (Fig. 1).

During March, ca. 11% of eggs tested hatched at the 1st flooding and followed by 47, 24, and 17% of hatch at each subsequent flooding (Table 3). However, in order to obtain a 100% hatching of the viable eggs in the samples tested, at least 10 separate floodings were required. Temperature in this period varied 70° to 40°F (21.1° - 4.4°C) and daily sunshine increased from 6 hrs to 9.5 hrs (Fig. 1).

During April and May, ca. 51.5% of eggs tested hatched at the 1st flooding, but 11 separate floodings were required to get the perfect hatch. During this period, temperature varied 85° to 42°F (29.4° - 5.6°C) and photoperiod was 9.5 to 12.5 hrs.

The most unexpected hatching pattern was obtained from results of the tests conducted during October; 12.4% at the 1st, 11.6% at the 2nd, 2.3% at the 3rd, and then up to 20% at the 4th, then down to 6.3% at the 5th flooding (Table 4, Fig. 1). During this period, temperature fluctuated 85° to 40°F (29.4° - 4.4°C) and daily photoperiod reduced from 11.5 to 7.8 hrs. Egg diapause is an important survival

Table 1.—Percent hatch at each flooding of *Aedes nigromaculis* eggs collected from field during June to September.

Flood- ing	Date of 1st flooding							Mean
	June 20	July 7	July 30	Aug. 4	Aug. 20	Sept. 8	Sept. 14	
1st	92.68	93.51	94.63	94.98	90.68	93.81	95.09	93.62
2nd	6.12	5.19	4.13	3.56	6.83	6.02	4.40	5.18
3rd	1.30	1.30	1.24	1.46	2.48	.13	.50	1.20
4th	0	0	0	0	0	.04	0	.01
5th	0	0	0	0	0	0	0	0
No. eggs	625	77	242	618	161	2,343	1,386	5,452

Table 2.—Percent hatch at each flooding of *Aedes nigromaculis* eggs collected from field during November, December, and January.

Flooding	Date of 1st flooding			Mean
	Jan. 12	Nov. 5	Dec. 9	
1st	0	.05	0	0.02
2nd	27.21	56.07	22.56	35.28
3rd	64.54	18.24	4.18	28.99
4th	3.37	12.94	73.02	29.78
5th	0	8.63	0	2.88
6th	.98	3.69	.16	1.61
7th	2.48	.14	.08	.90
8th	.75	.12	0	.30
9th	.58	.02	0	.20
10th	0	.10	0	.03
11th	.09	0	-	.03
12th	0	0	-	0
13th	0	-	-	0
No. eggs	2,253	4,173	2,465	8,891

mechanism for the pasture mosquito during winter periods; it probably begins long before the onset of winter conditions (Table 4, Fig. 1) and it may not be terminated completely until long after the vanishing of winter conditions (Table 3, Fig. 1).

Table 5 shows the results of a preliminary test on termination of egg diapause. The eggs exposed to a "14L:10D,<sup>1</sup> 80° - 60° F" for 49 days completely terminated diapause. The eggs exposed to a "24D, 80° - 60° F" for the same time did not hatch at the 1st flooding, but 99% hatched at the 2nd flooding showing some temperature influence on termination of diapause. Hatching test of Control 1 was made at the time when the bunch grass clump was collected from the field (September 6); Control 2 was performed with the same bunch grass used for Control 1, but stored outdoors

<sup>1</sup>L : hours of light, D : hours of darkness.

Table 3.—Percent hatch at each flooding of *Aedes nigromaculis* eggs collected from field during March.

Flooding	Date of 1st flooding		Mean
	March 6	March 30	
1st	5.73	16.02	10.88
2nd	13.40	80.77	47.08
3rd	46.42	2.26	24.34
4th	33.42	.68	17.05
5th	.74	0	.37
6th	.10	.11	.10
7th	.15	0	.08
8th	0	.06	.03
9th	.05	.06	.03
10th	0	.06	.03
11th	0	0	0
12th	-	0	0
No. eggs	2,023	1,773	3,796

for 6 months. The results of hatching tests of the controls also indicate the direct effect of the environmental factors upon embryonic diapause.

According to Beck (1968), embryonic diapause of insects (lymantrid moths, silk worms, aphids, and mites) is usually caused by the photoperiods experienced by the parent generation rather than eggs themselves. However, in case of the pasture mosquito egg diapause, it seems that temperature and photoperiod play an important role in the induction and termination of diapause, acting either on the parent generation (Miura et al. 1968, McHaffey 1972) or directly on the egg.

#### SUMMARY.—

1. *A. nigromaculis* eggs collected from breeding sites during June to September hatched readily at the 1st flooding.
2. Eggs collected during November to January did not hatch at the 1st flooding, but hatched 35% at the 2nd, 29% at the 3rd, 29% at the 4th, and 3% at the 5th flooding.

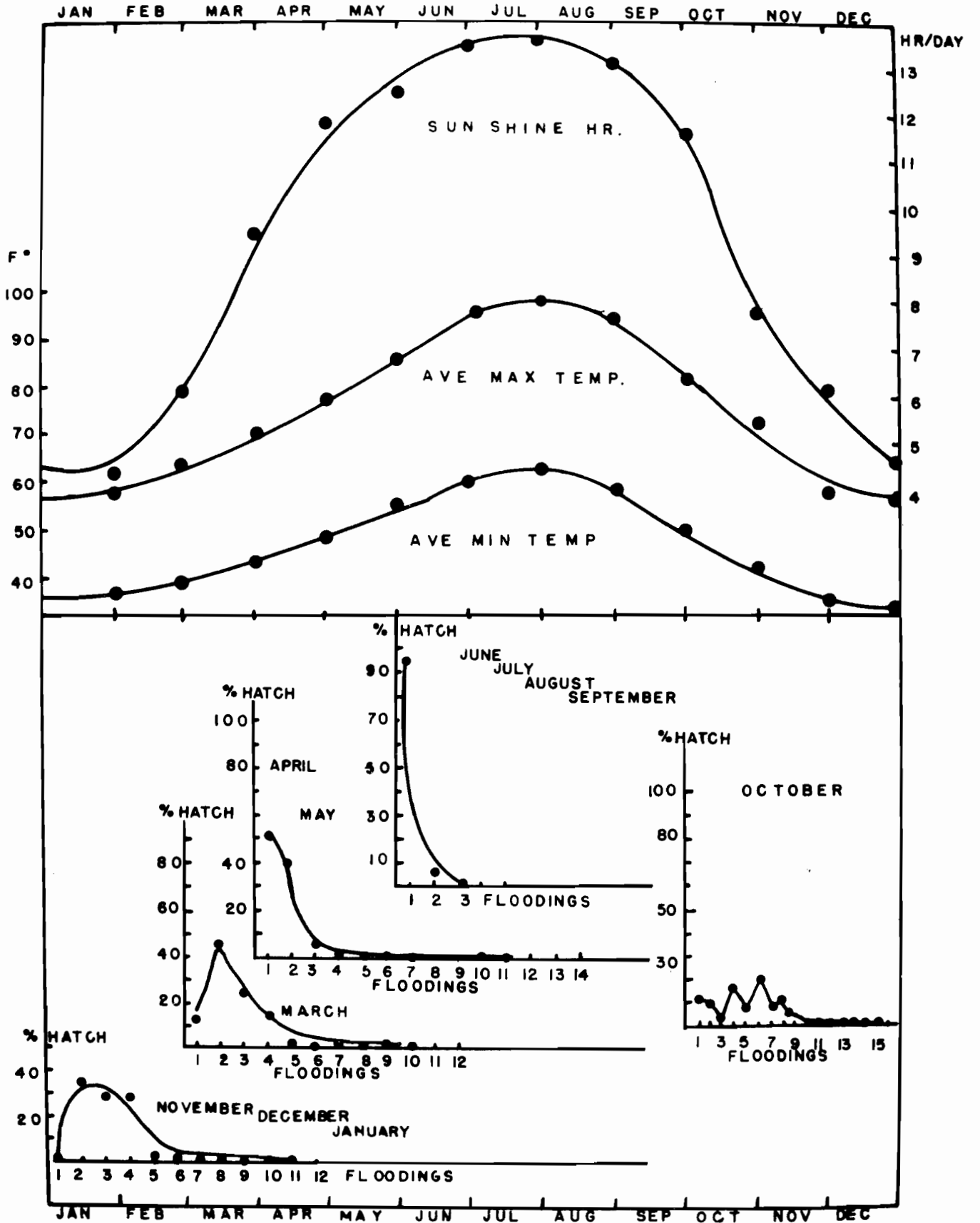


Fig. 1.—Seasonal hatching variation of *Aedes nigromaculis* eggs together with data showing the average maximum, minimum temperature and daily photoperiod.

Table 4.—Percent hatch at each flooding of *Aedes nigromaculis* eggs collected from field during April and May.

Flooding	Date of 1st flooding			Mean
	April 26	May 10	May 17	
1st	72.37	53.07	8.13	51.48
2nd	15.40	42.52	89.96	40.82
3rd	6.66	3.13	.85	4.48
4th	1.22	.44	.91	.95
5th	.24	.24	.15	.22
6th	.49	0	0	.25
7th	.73	0	0	.36
8th	.73	-	-	.36
9th	0	-	-	0
10th	0	-	-	0
11th	1.22	-	-	.60
12th	.98	-	-	.50
13th	0	-	-	0
No. eggs	409	2,521	3,298	6,228

- Eggs collected during March hatched 11% at the 1st flooding, 45% at the 2nd, 25% at the 3rd, and 17% at the 4th flooding.
- Eggs collected during April and May hatched 52% at the 1st flooding, 41% at the 2nd, 5% at the 3rd, and 1% at the 4th flooding. In order to get 100% hatch, 10 to 12 separate floodings are required.
- Eggs collected during October hatched in the most unexpected hatching pattern; 12.5% at the 1st, 12% at the 2nd, 2% at the 3rd, 17% at the 4th, and 6% at the 5th flooding.
- The diapaused eggs exposed to a "14L:10D, 80° - 60°F" condition for 49 days terminated diapause.
- The diapaused eggs exposed to a "14D, 80° - 60°F" condition for 49 days showed some reactivation but needed the 2nd flooding to obtain 100% hatch.
- Speculation is made that eggs of *A. nigromaculis* enter diapause probably in response to descending temperature

Table 6.—Results of preliminary study of termination of diapaused *Aedes nigromaculis* eggs. (Bunch grass clumps were collected on September 8, 1971, and stored outdoors at the laboratory.)

Treatment (Jan. 12 to March 1)	Number eggs	% hatch at each flooding						
		1st	2nd	3rd	4th	5th	6th	7th
14L:10D, 80° - 60°F <sup>1</sup>	942	99.04	0.64	0.11	0	0.11	0.11	0
24D, 80° - 60°F <sup>2</sup>	8,081	0	99.00	.64	.26	.10	0	0
Control 1 <sup>3</sup>	3,151	95.27	4.70	.03	0	0	-	-
Control 2 <sup>4</sup>	147	6.12	3.40	26.53	60.54	2.04	0	1.36

<sup>1</sup> A daily cycle of 14 hr light and 10 hr darkness with temperature fluctuating 80° to 60°F (26.7° - 15.6°C); 1st flooding on March 6, 1972.

<sup>2</sup> A daily cycle of 24 hr darkness with temperature fluctuating 80° to 60°F; 1st flooding on March 6, 1972.

<sup>3</sup> 1st flooding on September 14, 1971.

<sup>4</sup> 1st flooding on March 6, 1972, kept in natural condition; no artificial treatment.

Table 5.—Percent hatch at each flooding of *Aedes nigromaculis* eggs collected from field during October.

Flooding	Date of 1st flooding					Mean
	Oct. 6	Oct. 15	Oct. 19	Oct. 22	Oct. 29	
1st	15.11	34.15	6.13	3.80	3.03	12.44
2nd	24.18	15.28	1.74	4.44	12.62	11.65
3rd	.64	.45	2.38	4.18	3.88	2.31
4th	.16	.76	10.63	7.98	78.37	19.58
5th	1.54	2.71	9.73	16.47	1.01	6.29
6th	.90	41.38	10.81	51.54	.90	21.11
7th	17.34	4.68	7.48	7.37	0	7.37
8th	19.41	.47	29.26	3.51	.20	10.57
9th	8.80	.05	18.23	.06	0	5.43
10th	10.71	.06	.31	.29	0	2.27
11th	.10	0	.11	.20	-	.08
12th	6.36	.01	2.23	.17	-	1.75
13th	1.00	0	.31	0	-	.26
14th	.10	0	.51	0	-	.12
15th	0	0	.13	0	-	.03
No. eggs	1,886	48,337	9,137	3,448	4,152	66,960

and photoperiod during October; and terminated diapause in response to ascending temperature and photoperiod during March.

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# BIOLOGICAL CONTROL OF AQUATIC WEEDS IN THE LOWER COLORADO RIVER BASIN

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Increasing costs of labor and herbicides plus increasing awareness of the threat of pollution from herbicides, pesticides, and nutrients from agricultural runoff are complicating aquatic weed (= "moss") control programs in irrigation and water districts of the lower Colorado River basin. Canals, drains and tiles variously become choked or clogged with aquatic vegetation, and require continued maintenance for the successful operation of the extensive irrigation and drainage system. Control methods vary within different areas of the basin and in general are experiencing an unacceptable increase in control/cost ratio. Hopefully, control would ultimately make maximum use of natural enemies (phytophagous fish, snails and disease pathogens), physical habitat manipulation to disfavor plant growth and enhance the activity of weed-feeders and a more selective integration of currently available or to-be-developed herbicides.

Chemical control with known materials and formulations is difficult and increasingly expensive because of the dilution factor and the neutralizing effect that waters of high organic content have upon many compounds, and the wide dispersal of weeds through the irrigation system. Also recognized is the potential danger to aquatic dependent fauna in the Salton Sea deltas and Colorado River that is posed by runoff of herbicides and other pollutants.

The best known natural enemies of aquatic weeds are fish and phytophagous snails, which are presently difficult to maintain over the greater area of the Colorado River basin without some type of habitat modification that would assure natural breeding and winter survival. Also, whereas higher aquatic weeds and algae in stable habitats characteristically reduce and limit their population density after a period of time through antimetabolite pollution and nutrient exhaustion, those in the Colorado basin drainages inhabit water that is constantly cleansed and resupplied with a stream of new nutrients. The inevitable consequence is apparent: the potential for prolonged nuisance of aquatic vegetation in the basin is substantially great.

**CURRENT APPROACHES AND RESULTS: Weed Survey.**—A survey of aquatic weed species begun in 1970 is being continued on a regular basis to determine the number of species concerned and the seasonal incidence of each. Information from this survey furnishes a guide to the type of control measures and species of weed feeders of greatest potential. Also, these data identify those weed species that are actually creating problems and those that can be at least temporarily disregarded, thus improving the efficiency of the research effort. The principal weed species as assessed from completed surveys include at least 3 species of *Potamogeton*, *Elodea canadensis* Michx., and several species of filamentous green algae: (*Cladophora* spp., etc.)

and *Chara* spp. Of more limited problem status are emergent weeds such as cattails (*Typha* spp.) and bulrushes (*Scirpus* spp.) (see Otto and Bartley 1965 for descriptions and illustrations of weeds concerned).

**Weed Feeders.**—Candidate weed feeders proposed for study in paved laterals, ditches, and drainage areas include 8 species representing 4 genera of fish: 3 species of *Tilapia*, 3 species of *Mollienisia*, 1 *Cyprinodon* and the grass carp, *Ctenopharyngodon idellus*.

*Tilapia* are prolific fast growing African cichlid fish that have been transported to several parts of the world, including the State of Hawaii, for noxious aquatic weed consumption. Their additional value in biological control of mosquitoes comes about through their feeding activities on emergent vegetation and algal mats, thereby eliminating protective niches for mosquitoes (Legner and Medved 1972). We have studied 3 species, *T. mossambica* (Peters), *T. zillii* (Gervais), and *T. hornorum* Trewazas. Although winterkill reduces their populations, we have found that given the right breeding conditions reproduction is rapid enough in the spring to produce adequate numbers for summer and fall algae and other aquatic plant control. The species mentioned have performed remarkably in ponds at the University of California, Riverside (Figs. 1 and 2), and in certain test drains in the Imperial Valley, even to the extent of significantly curtailing the growth of cattails (*Typha* spp.)

The *Mollienisia*, or mollies, are small, attractively patterned Central and South American poeciliid fish that are by nature largely vegetarian. Being very active, they eat often, especially in the manner of nibbling at algae, which they do almost constantly. We have observed 3 species, *Mollienisia latipinna* LeSueur, *M. sphenops* (Cuvier and Valenciennes), and *M. sp.* at Riverside. Besides their potential benefits as algae feeders, these beautifully colored fish feed on larvae of midges and mosquitoes. Among their favorable attributes is a rather wide range of tolerance to solute concentration and temperatures.

The desert pupfish, *Cyprinodon macularis* (Baird and Girard), already inhabiting certain portions of the lower Colorado River basin, has desirable qualities as an algae feeder. Techniques to increase the range of this small native species might be devised for the purpose of both algae and mosquito control.

The phytophagous habits of certain aquatic snails, e.g., *Marisa conrvarietis* (L.), has been demonstrated to practically reduce aquatic plant growth in recent tests at Riverside. At least 2 species of snails have been accidentally introduced into the area around the Salton Sea but their role in "moss" control is yet to be determined.

Studies have shown it to be a useful species for consuming enormous amounts of diverse kinds of aquatic vegeta-

tion without any harmful effects on crop plants such as rice. These observations plus the knowledge that *C. idellus* will not reproduce naturally in America (personal communication, A. J. Calhoun) has enabled us to consider its importation on a quarantined basis into portions of the lower Colorado River basin.

**Fish Behavior and Management.**—From our data gathered since 1971, there appears to be a high degree of intercompatibility among all species of fish studied thus far including *Gambusia*, the mosquitofish, so that the establishment of a complex of desirable species throughout the basin is conceivable. Territorial behavior of *Tilapia* especially, poses a recognized problem to practical biological control. In order for these fish to be effective in reducing aquatic plant growth, a minimum density per unit of area must be maintained. In controlled habitats, adequate densities are generally attained during the first year of establishment where water temperatures exceed 22°C, during which time the growth, a minimum density per unit of area must be maintained. In controlled habitats, adequate densities are generally attained during the first year of establishment where water temperatures exceed 22°C, during which time the aquatic environment consistently becomes remarkably cleansed of noxious plant growth (Legner and Medved 1972; Figs. 1 and 2; Legner, unpub. data). However, observations in test plots in the Imperial Valley since 1970 and in Riverside since 1971 show that dominant male *Tilapia* begin to stake out territories of varying diameter—generally exceeding 10 meters, within which only a few females are tolerated. Resulting fry are either driven out of the territory or cannibalized. This results in a thinly dispersed *Tilapia* population which is unable to keep ahead of the build-up of aquatic weeds. Such territorial behavior

appears to be interspecific and does, for instance, noticeably affect cohabitation with a newly planted population of another species (e.g. *T. zillii*). Ways to interrupt and/or alter this behavior seems to be essential to acquire the fish density necessary for maximum aquatic weed reduction. Current studies are designed to devise techniques that would effectively rogue out the dominant males as they appear through the use of angling, trapping, shooting, and pheromones. In an aquatic area as large as that of the lower Colorado River basin, all 4 of the practices singly or in combination might be necessary for proper management. The greatest problem with the effective use of the white amur, *C. idellus*, is availability. Before its use as an aquatic weed control agent can be effectively implemented, a mass production technique developed from basic nutritional studies will be required.

Environmental impact of the introductions of new weed consuming species in the man-made and sustained irrigation system of the lower Colorado River basin is being carefully considered and studied both as intensely as our support funds permit, and in close cooperation with biologists of the California Department of Fish and Game. Any adverse effects are already expected to be minimal compared to the overall benefits derived from the reduction of herbicide usage and labor costs. Therefore, it is certain that through the use of inexpensive non-polluting biological control organisms, the water quality can be expected to improve. As a result, this country's commitment to the Mexican government to supply better quality water from the Colorado River drainage system can be more readily realized. Also, the widespread distribution of fish, such as the *Tilapia*, that are capable of reducing mosquito-breeding habitats undoubtedly will reduce significantly the threat of invading mosquito-carried viruses such as Venezuelan equine encephalitis.



Fig. 1.—Effects of 2 male and 3 female, 1 month-old *Tilapia mossambica* after feeding for 1 week on aquatic vegetation in 2 sections of a divided shallow pond (foreground) at Riverside, Calif. at 23°C. The center section of this replicate is the check (no fish). Floating filamentous algae and submerged aquatic plants were significantly reduced in three replicated ponds.



Fig. 2.—Effects of 2 male and 3 female, 6 weeks-old *Tilapia zillii* after feeding for 3 weeks on the roots of cat-tails (*Typha* sp.) and other aquatic vegetation in one section of a divided shallow pond (foreground) at Riverside, Calif. at 23-25°C. Only the center section contains the fish. Floating filamentous algae and submerged aquatic plants were also significantly reduced in three replicated ponds.



### Acknowledgment

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## CURRENT PROGRESS ON *BEAUVERIA TENELLA* AS A CONTROL AGENT FOR MOSQUITOES

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The fungus *Beauveria tenella* was isolated from "sick" larvae of the tree hole mosquito *Aedes sierrensis* from Marin County, California (Sanders 1972). Laboratory tests have shown that this fungus is pathogenic for seven species from three genera of mosquitoes, namely the host from which it was isolated *A. sierrensis*, and *A. dorsalis*, *A. aegypti*, *Culex pipiens*, *C. tarsalis* and *Culiseta incidens* (Pinnock et al. 1973). The fact that the fungus was pathogenic to all the species tested lends encouragement as to its potential pathogenicity for other genera and species as well.

*A. sierrensis* and *C. tarsalis* did not appear to be as susceptible to the blastospores of *B. tenella* as the other species. All tests referred to here were carried out at 12-14° C. At  $5 \times 10^7$  spores per ml *C. tarsalis* showed 100% mortality after 11 days while at lower concentrations ranging from  $5 \times 10^4$  to  $5 \times 10^6$  complete mortality was not achieved until 21 days. *A. sierrensis* reached 100% mortality at all concentrations only after 21 days. For this latter species there was essentially no difference in the rate of mortality at any of the concentrations. All species with the exception of *A. sierrensis* showed an increase in the rate of mortality at  $5 \times 10^7$  spores per ml generally reaching 100% mortality between 4 and 11 days.

Early instar mosquitoes appeared to be more susceptible than later instars. In tests with *A. aegypti*, 1st instar larvae reached complete mortality after 4 days while only  $\frac{3}{4}$  of the 2nd instars and  $\frac{2}{5}$  of the 4th instars of this species died during the same period.

Field tests against *A. sierrensis* in tree holes using dosages of  $5 \times 10^3$  and  $5 \times 10^5$  spores per ml caused a reduced emergence of adult mosquitoes. At the high dosage less than 30% of the expected adults emerged while at the low dosage the emergence was about 50%. Further field tests are being conducted and planned against this species and several others during the current mosquito season.

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# COMPARISON OF JUVENILE HORMONE ANALOGUE FORMULATIONS AGAINST CHIRONOMID MIDGES UNDER SEMI-FIELD CONDITIONS

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## ABSTRACT

Laboratory assay showed emergence of *Chironomus* *stigmaterus* and *Tanytus* *grodhausi* collected from sewage ponds as 4th instar larvae to be completely inhibited by Altosid (ZR-515) at .05 ppm. Larval mortality occurred proportionate to dosage. In outdoor box plots (3ft x 3ft x 10 in) members of the subfamily Chironominae always re-

covered more rapidly (1 wk) from treatment than did the Tanypodinae (2 wks). A 5% heavy flowable formulation (microencapsulated) of Altosid exhibited slightly longer residual activity than either the 10% light flowable or the 4 lb/gal EC. No difference could be detected between the activity of the 10% and the EC materials.

PREDATION OF MOSQUITOES AND CHIRONOMID MIDGES IN PONDS BY  
*TILAPIA ZILLII* (GERVAIS) AND *T. MOSSAMBICA* (PETERS) (TELEOSTEI: CICHLIDAE)

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The quest for suitable biological control substitutes for *Gambusia affinis affinis* (Baird and Girard) on mosquitoes and *Cyprinus carpio* L. on chironomid midges, has focused attention on the cichlid genus *Tilapia*. These fish feed on chironomid midges and mosquitoes (Hallock and Ziebell 1970, St. Amant 1966, J. A. St. Amant, personal communication); and certain species are capable of significantly reducing mosquito breeding habitats through grazing on aquatic vegetation (Hickling 1961, Legner and Medved 1972, Legner et al. 1973). Two species, *Tilapia mossambica* (Peters) and *T. zillii* (Gervais) were investigated for the effectiveness in reducing pond populations of mosquitoes and chironomid midges at Riverside, California during 1972. Preliminary results of these studies are reported herein.

**METHODS AND MATERIALS.**—Experimental ponds to investigate fish predation and behavior were constructed at the Aquatic Research Laboratory, University of California, Riverside (Legner and Medved 1972). Two types of ponds employed were earthen excavations, the first type, 5.5 m wide by 7.6 m long and 0.4 m deep, was subdivided into 3 subreplicates with barriers constructed of 11 mesh/cm saran screening (Fig. 1). The second type, 1 meter-square and 0.2 m deep, was enclosed with redwood frames (Fig. 2). Water from deep wells was continuously added from the University Experiment Station irrigation system to replace that lost by percolation and evaporation. Water quality varied with agricultural practices as water was recirculated through the experiment station area.

Groups of 2 male and 3 female, 6-week-old *T. zillii* and *T. mossambica*, were separately introduced into each of 3 random subreplicates of the larger ponds on March 9, 1972, and allowed to reproduce until cold-induced mortality occurred in late October. The progeny were allowed to remain in the replicates. One 6-week-old *T. mossambica* female was introduced into each of 6 redwood-frame ponds selected at random and left there until death without reproduction.

Twelve grams of chick-start feeding mash were applied weekly to the smaller meter-square ponds to stimulate continuous chironomid midge production. Nothing was added to the large ponds. Mosquito breeding in the smaller ponds gradually ceased as blood meals could not be attained.

Dip and dredge samples were taken periodically from all experimental units to estimate densities of mosquito and chironomid larvae. For mosquitoes, a standard 500 ml dipper was used in the large ponds with 4 dips being taken at random per each section every 7 days. Only 4th instars were counted and all stages were replaced in each replicate. The small ponds were sampled with a 40 ml dipper, 4 random dips per replicate every 7 days. For chironomids, a 250 ml benthos scoop dredge was used in the large ponds

with 3 scoops being taken at random per each section every 14 days. The small ponds were sampled with a 120 ml dredge, 1 scoop at random every 7 days. In addition, adult mosquitoes and chironomids were collected daily from the meter-square ponds in screened receptacles placed over each replicate (Fig. 2). Detailed observations also were made



Fig. 1.—Large subdivided experimental ponds, 5.5 x 7.6 m and 0.4 m deep, used to investigate fish behavior and predation at the Aquatic Research Laboratory, University of California, Riverside.

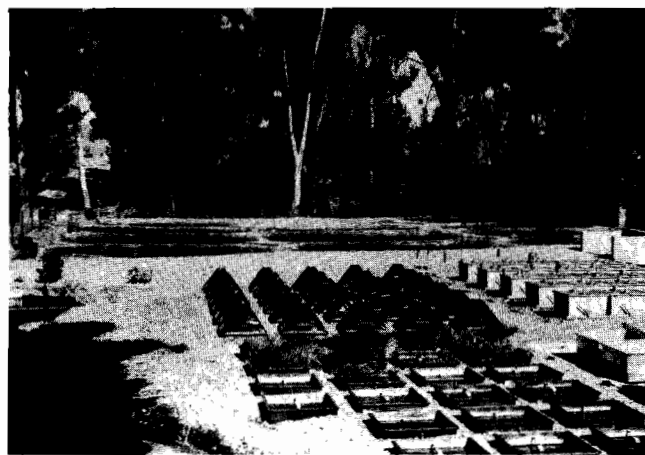


Fig. 2.—Small redwood-framed ponds, 1 meter-square and 0.2 m deep and emergence traps to collect adult mosquitoes and chironomids (background) for studying the behavior and predation of single female fish at the Aquatic Research Laboratory, University of California, Riverside.

Table 1.—Effect of *Tilapia* on the density of 4th instar mosquitoes in large ponds during the period April 17 - August 14, 1972, Riverside, California.

<i>Tilapia zillii</i>			<i>Tilapia mossambica</i>		
Average No. per four 500-ml dips			Average No. per four 500-ml dips		
Treatment	Check	% Reduction <sup>1</sup>	Treatment	Check	% Reduction
0.47 <sup>2</sup>	2.64	82.24	1.16 <sup>2</sup>	4.59	74.67

<sup>1</sup>100 - (treatment/check x 100)

<sup>2</sup>significantly lower than check at t<sub>01</sub>

Table 2.—Effect of *Tilapia mossambica* on the density of 4th instar mosquitoes and adult emergence in meter-square ponds during the period June 5 - July 3, 1972, Riverside, California.

Average No. larvae per four 40-ml dips			Average No. adults emerged/day		
Treatment	Check	% Reduction <sup>1</sup>	Treatment	Check	% Reduction
1.97 <sup>2</sup>	2.73	27.84	13.95 <sup>2</sup>	18.79	25.73

<sup>1</sup>100 - (treatment/check x 100)

<sup>2</sup>significantly lower than check at t<sub>05</sub>

on water temperature and fish growth, but only gross results on predation and interference with insect breeding are presented here.

**RESULTS AND DISCUSSION: Mosquitoes and Midges Present.**—The principal mosquito species and their relative abundance (which emerged from the check ponds during the experimental period from April through August) were *Culex peus* Speiser (50.6%), *C. tarsalis* Coquillet (35.6%), *C. pipiens quinquefasciatus* (Say) (12.7%), and *C. incidens* (Thomson) (1.7%). The principal chironomid species and their relative abundance were *Chironomus* spp., *Dicrotendipes* sp. *Procladius* sp. (combined 74.2%), *Tanytarsus* spp. (19.3%), and *Microspectra nigripila* (Johannsen) (6.5%).

**Fish Influences on Mosquito Density.**—Both species of *Tilapia* exhibited a pronounced significant suppression of mosquito larval density and adult emergence in both the large and small experimental ponds (Tables 1 and 2). Their performance in the large ponds was superior to the more restricted meter-square units. This was probably due to the more natural aspect of a larger area and the presence of fry. Schooling behavior was also possible in the large ponds, the fish traveling in groups of 10-25 individuals as progeny were added to the system by reproduction. These schools patrolled most of their habitat area, consuming mosquito larvae and pupae which they encountered.

The percent reduction of adult mosquitoes in meter-square replicates with fish was not significantly different from that estimated by dip samples, indicating the reliability of the larval sample for estimating emergence in this experiment. Although mosquito emergence from the large ponds could not be measured accurately, results from the

meter-square replicates indicate that it was probably similar to that estimated from larval dips. One hundred percent control of mosquitoes was achieved in all the experimental ponds after July 10 with *T. zillii* and July 31 with *T. mossambica*.

Reduction of mosquito densities by the fish was affected almost entirely by predation, there being apparently little disruption of the breeding habitat that would render it unsuitable for development of mosquito species in the area. Although mature fish were effective mosquito predators, the young fry appearing in July were the most effective. They eliminated all breeding about 2 weeks after their initial appearance. The fish also adversely affected the mosquito population by destroying the floating filamentous algae which provide protective niches for mosquito larvae and pupae (Legner and Medved 1972, Legner et al. 1973).

**Fish Influences on Chironomid Density.**—A significant suppression of chironomid larvae and their adult emergence also was observed with both *Tilapia* species, *T. zillii* being the most effective in control (Tables 3 and 4). However, the reduction of all emerging chironomid species was only half as great as that initially measured with the benthos larval sample (Table 4). This was probably due to selective feeding and disruption of larval habitats.

A continuing examination of chironomid species sampled in the experiments indicates the presence of at least 7 species or through the habitat disruption of others. Additional studies are being made to describe the species and to elucidate the behavioral characteristics of those emerging to better assess the importance of fish in abating chironomid nuisances.

Table 3.—Effect of *Tilapia* on the density of benthos chironomid larvae in large ponds during the period May 25 - August 3, 1972. Riverside, California.

<i>Tilapia zillii</i>			<i>Tilapia mossambica</i>		
Average No. per 250-ml scoop			Average No. per 250-ml scoop		
Treatment	Check	% Reduction <sup>1</sup>	Treatment	Check	% Reduction
2.97 <sup>2</sup>	4.82	38.36	3.13 <sup>2</sup>	6.01	48.01

<sup>1</sup>100 - (treatment/check x 100)

<sup>2</sup>significantly lower than check at t<sub>05</sub>

Table 4.—Effect of *Tilapia* on chironomid larval density and adult emergence in meter-square ponds during the period June 5 - September 19, 1972. Riverside, California.

<i>Tilapia zillii</i>			<i>Tilapia mossambica</i>			<i>Tilapia zillii</i>			<i>Tilapia mossambica</i>		
Ave. No. per 40-ml scoop			Ave. No. per 40-ml scoop			Ave. No. adults emerged/day			Ave. No. adults emerged/day		
Treat-ment	Check	% reduction <sup>1</sup>	Treat-ment	Check	% reduction	Treat-ment	Check	% reduction	Treat-ment	Check	% reduction
5.85 <sup>2</sup>	18.00	67.50	17.16 <sup>2</sup>	38.72	55.68	96.21 <sup>2</sup>	138.29	30.43	183.55 <sup>2</sup>	262.10	29.97

<sup>1</sup>100 - (treatment/check x 100)

<sup>2</sup>significantly lower than check at t<sub>01</sub>

Fish acted to reduce chironomid larvae through direct predation in the benthic area of the ponds and by habitat disruption. Fish nudging in the benthos with their mouths consumed midge larvae and disturbed adjacent tube-forming species, causing mortality in some cases.

Outlook for Biological Control.—The *Tilapia* may be useful candidates for integrated mosquito and chironomid midge control, their usefulness varying with water temperatures and characteristics of the breeding habitat.

The age structure of fish populations apparently greatly influences the degree of control attainable with these species. In field establishment trials in irrigation drains of the lower Colorado River basin, the dominant male *Tilapia*, appearing in one-year old stock were observed to demonstrate strong territorial behavior which reduces a species' density. Means to interrupt this behavioral trait to increase the density of mosquito and chironomid-feeding *Tilapia* are currently being investigated.

Environmental impact studies have shown that populations of *T. mossambica* decline rapidly in water below 12°C (St. Amant 1960) and *T. zillii*, below 7°C (Legner, unpub. data), which greatly reduces field populations annually. Though prolific breeders in water warmer than 22°C, they sustain a significant annual dieback, eliminating any tendency toward overpopulation as observed in southeast Asian populations of *T. hornorum* Trewazas (Hickling 1961). We also have observed that some predation by predatory fish such as bass further reduces the number of fry in summer populations.

Both *T. mossambica* and *T. zillii* are good game fish, responding favorably to angling enthusiasts. The edible qualities of young adults in the size range of 16-30 cm are desirable. Thus, their addition to waters of the lower Colorado River basin would be harmonious with fisheries improvement goals and environmental aspirations.

#### Acknowledgment

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# LABORATORY OBSERVATIONS AND FIELD TESTS WITH *LAGENIDIUM* AGAINST CALIFORNIA MOSQUITOES

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**INTRODUCTION.**—In 1969, C. J. Umphlett and C. S. Huang isolated an aquatic fungus from infected *Culex* larvae collected in Orange County, North Carolina, and provisionally named it *Lagenidium culicidum* (Umphlett and Huang 1970). Recent taxonomic revision indicated the organism to be a strain of *Lagenidium giganteum* (Couch) (Umphlett 1973). Studies at the Technical Development Laboratories in Savannah during the past three years (McCray et al. 1973) indicated that the organism was an excellent candidate for further evaluation as a potential biologic control tool.

Numerous factors including life cycle, culture methods, susceptible mosquito species, environmental limitations, dosage rates, effects on selected nontarget organisms and methods of application have now been elucidated, and analysis of these data revealed the organism to have an excellent potential for the control of several culicine species including *Culex tarsalis*, the primary vector of encephalitis in California.

Limited exploratory field studies were conducted during the latter part of the summer of 1972, with the cooperation of the California Bureau of Vector Control and Solid Waste Management. The objectives were: to determine whether a single introduction of this strain of *Lagenidium* into selected sites would result in infection and reduction of the natural population; to study those environmental factors that might influence the use of this aquatic fungus as a biologic control organism; and to make observations on the aquatic nontarget organisms present that might be affected by the introduction of this agent.

This paper reports additional laboratory observations and the results of the field studies conducted in a rice field environment against *C. tarsalis* north of Sacramento near Colusa and in irrigated pastures against *Aedes nigromaculis* south of Fresno near Hanford.

**MATERIALS AND METHODS.**—1. Site Selection and Description.—The area near Colusa was primarily one of rice fields. Adjacent to the rice fields were irrigation canals and drainage ditches used to control irrigation water. The rice field and drainage ditches which returned the water to the irrigation system. The rice fields and drainage ditches appeared to be the primary mosquito breeding sites. However, because of our limited knowledge of the effect of the test organism on nontarget species, site selection was restricted

to small, "dead end" bodies of water that did not flow into ditches, drainage canals or streams that might spread the *Lagenidium* throughout the environment. Another restriction was that the experimental sites be such that shortly after the termination of the test, the sites would dry up.

Three test sites meeting these requirements were selected near Colusa. Each of the sites were seepage ditches adjacent to rice fields. Two were divided with small dams or dikes into three test plots approximately 3 x 30 feet long, and one was a single, undivided test plot 90 feet long. Each test site also had two control plots, either adjacent drainage ditches or portions of the seepage ditches themselves.

The water from each test and control plot and the adjacent rice fields was analyzed for total dissolved solids (TDS), pH, percent salinity and chloride ions. Water temperatures of each test and control plot were recorded daily.

These data revealed that the rice fields and drainage ditches, the principal habitat of *C. tarsalis*, had a pH of approximately 8.0, total dissolved solids of 190.7 mg/liter, chloride ions of 11.25 mg/liter and a percent salinity of 0.05. The data from the water analyses also revealed that of the three seepage ditches selected as test sites, only site no. 2 had water qualities like those of the rice fields and drainage ditches. Site no. 1 had a pH of 10.0, TDS of 6,247 mg/liter, chloride ion concentration of 380.0 mg/liter and a percent salinity of 0.72. All of these water qualities were considered detrimental to the fungus and on the basis of existing laboratory data should have reduced the effectiveness of the fungus to totally unsatisfactory levels. At site no. 3 the chloride ion concentration was about 25 times those of the normal *C. tarsalis* habitat and the site was solidly overgrown with salt grass.

On the basis of these data, site no. 2 was the only acceptable test site. However, in order to verify in the field the data obtained in the laboratory, sites nos. 1 and 3 were retained in the tests.

Near Hanford, south of Fresno, a satisfactory test site was located in the irrigated pasture lands. It contained numerous checks (subdivisions of an irrigated field bounded by a levee) which were being flooded on a regular cycle; was an active mosquito breeding situation; the irrigation system was so designed that no runoff water escaped into another drainage system; and the cattle were not being grazed on the land.

One check was selected as a test site and one immediately adjacent was used as a control. Four square wooden frames were constructed, each four feet long on each side and about seven inches high. Around the bottom of each was stapled a skirt of black cotton cloth. The day after the checks were flooded, three of the frames were dropped in the water in selected locations in the test check and one in the control check so that uniform sampling plots existed. The frames were forced into solid contact with the sod and

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# LIFE CYCLE OF *LAGENIDIUM GIGANTEUM*

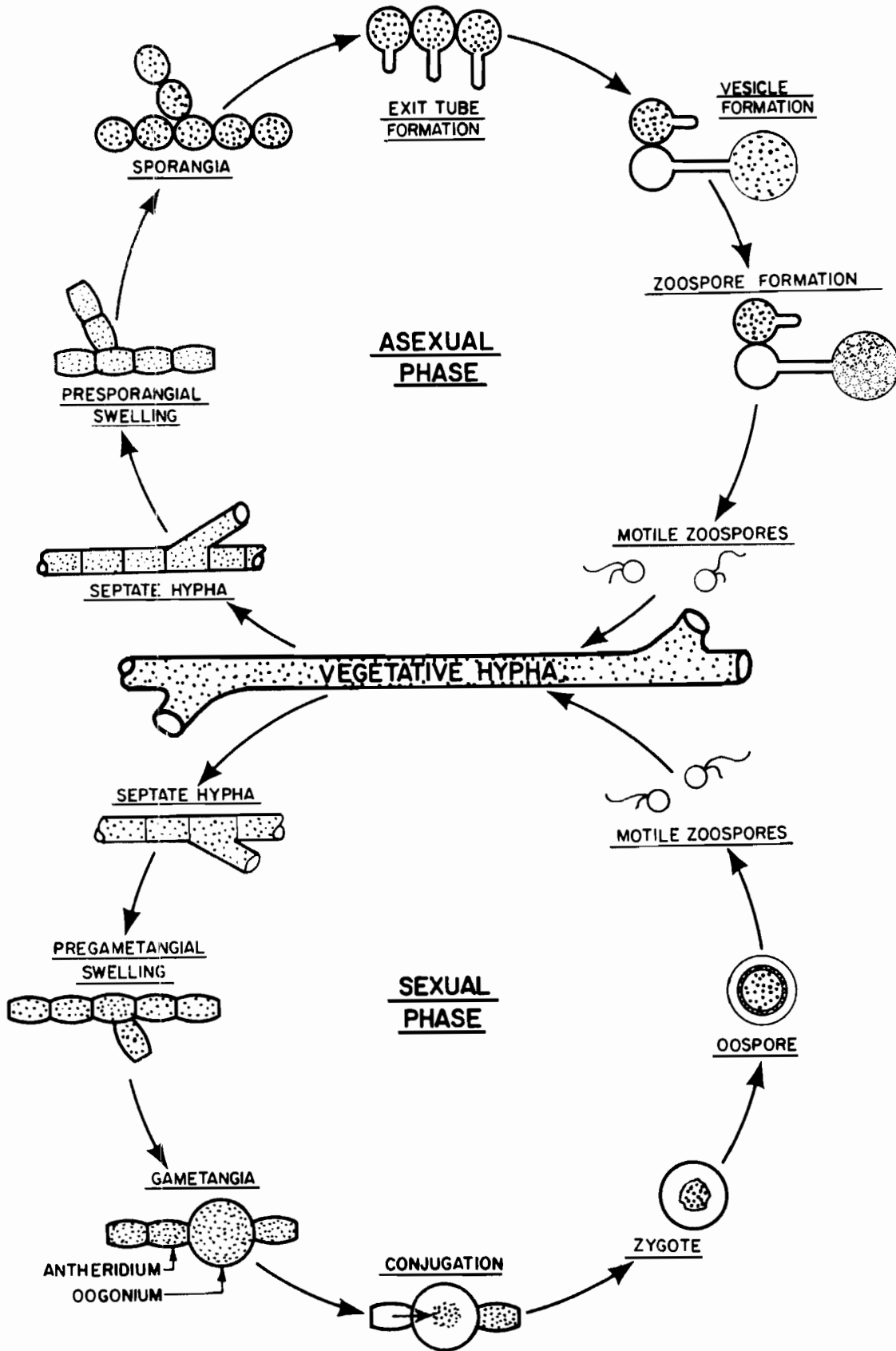


Fig. 1.-

Table 1.—The number of living *Culex tarsalis* larvae collected and found infected following introduction of *Lagenidium* at sites near Colusa, California, 1972.

Day after treatment	2	3	4	5	Total
Site No. 1					
Larvae collected	388	399	206	198	1191
Larvae infected	0	100	3	2	105
Percent infected	0	25.0	1.4	1.0	8.8
Site No. 2					
Larvae collected	146	101	8	0	255
Larvae infected	146	101	8	—	255
Percent infected	100	100	100	—	100
Site No. 3					
Larvae collected	114	81	45	46	286
Larvae infected	0	15	3	4	22
Percent infected	0	18.5	6.7	8.7	7.6

Table 2.—Mean daily pre- and post treatment collections of living *Culex tarsalis* larvae and pupae from Colusa site No. 2 inoculated with *Lagenidium*.

Day	-4	-3	-2	-1	0 <sup>1</sup>	+1	+2	+3	+4	+5	+17
Control <sup>2</sup>	110	122	102	125	88	72	93	80	123	112	111
Test <sup>3</sup>	96	96	78	93	89	88	51	36	5	0	0

<sup>1</sup>Day of inoculation.

<sup>2</sup>All instars from two plots combined.

<sup>3</sup>All instars from three plots combined.

Table 3.—The number of living *Aedes nigromaculis* larvae collected and found infected following introduction of *Lagenidium* at the site near Hanford, California, 1972.

Sampling plot	Day of treatment	3	4
Test 1	411	0	0
Test 2	321	3*	0
Test 3	309	0	0
Control 1	367	75	24

\*All three larvae died and were infected with *Lagenidium*.

held firmly in place with wooden stakes nailed to the sides. The cloth skirts were then pinned to the sod with nails in an effort to securely seal each sampling plot to prevent ingress or egress of larvae. Detailed water analyses were not made at this site, but pH ranged from 8.3 to 8.7 and the water temperature ranged from 79 to 82°F — all satisfactory qualities.

2.—Pretreatment Evaluations.—Population densities were determined in the Colusa area by taking five dips in each test and control plot, each at about 5-foot intervals along the seepage ditch. At the Hanford site the density was determined in each sampling plot by taking five dips, one in each corner and one in the center.

Pretreatment studies of each test and control plot consisted of determination of larval density; identification of species present; collection of nontarget aquatic organisms; and examination of the collected mosquitoes and nontarget organisms for the presence of pre-existing *Lagenidium* or other fungi that might be confused with *Lagenidium*. *C. tarsalis* was the predominant species in the Colusa area and *A. nigromaculis* was the predominant one in the Hanford area. *Anopheles freeborni* were present in both locations and *C. tarsalis* appeared several days after the start of the test in the Hanford area. No *Lagenidium* or similar fungus was observed as parasites of the mosquito larvae or the nontarget organisms collected.

3.—Production of Inoculum and Site Inoculation.—Eggs of *Aedes aegypti* from the University of California at Davis were hatched and larvae reared to produce sufficient 3-day-



old larvae as culture material for producing the fungal inoculum. Viable zoospores were shipped from TDL for inoculating the host larvae. Twenty-four hours after the infected host larvae died, they were fragmented and poured through a fine mesh sieve to remove any large particles such as head capsules. The resultant suspension was then placed in a 3-gallon pressure spray can and, using an 8002 nozzle and 40 lb pressure, each test plot in the Colusa area was treated at the rate of four fragmented cadavers (potentially one million zoospores) per linear foot of ditch. At the Hanford site the lower end of the irrigated test check, about 2,000 square feet of standing water, and the three sampling plots were treated at the rate of one fragmented cadaver (potentially 250,000 zoospores) per square foot.

4.—Post treatment Evaluations.—At the Colusa site, beginning the day of treatment and daily thereafter for five days, each plot was sampled by taking the five standard dips and concentrating the material in a "gussie bucket" (Womeldorf et al. 1963). All collected material was returned to the laboratory and the larvae and nontarget organisms were separated. All dead larvae were examined for infection with *Lagenidium*. All living larvae were separated by instar, plot, date, counted and held for a minimum of four days. Daily mortality was recorded for each, and every larva that died was examined microscopically for infection with *Lagenidium*. As many nontarget organisms as possible were also examined microscopically for infection with *Lagenidium*.

At the Hanford site, on the day of treatment 100 field-collected 2nd stage larvae were brought to the laboratory and divided among four containers holding about 100 ml of distilled water and 10 ml of the inoculum used in treating the test site. Field evaluations were made by obtaining counts on the day of treatment from each of the sampling plots by using the five standard dips and again on the 3rd and 4th posttreatment day. All dead larvae collected were examined immediately for infection and all living larvae were held for a minimum of four days. All larvae that died were examined for *Lagenidium*.

## Results

### A. Laboratory Observations.

#### 1. Parasitic cycle in *C. tarsalis*.

##### a. Vegetative phase.

The infective form of *L. giganteum* is the biflagellate, motile zoospore, and there were two distinct portals of entry for the mosquito larvae; the mouth and the cuticle.

In the majority of the infected larvae the zoospores were ingested and penetrated the tissues of the host larvae in the anterior portion of the digestive tract, usually the region of the pharynx. Mycelial growth spread rapidly from the point of entry throughout the head, then via the haemocoel toward the posterior, infecting the anal segment and gills last.

In about 2% of the specimens observed, however, mycelial growth first appeared in the abdomen, anal segment or gills. In these larvae the hyphae grew from darkly pigmented spots on the chitinous

exoskeleton. Larvae were routinely observed with infections occurring through both portals of entry. The hyphae grew via the haemocoel and spread rapidly throughout the body consuming the entire contents except for the chitinous respiratory tract. The hyphae then became septate and entered their reproductive phase.

#### b. Asexual reproduction (Fig. 1).

The individual segments of the hyphae became rounded, granular structures known as sporangia and sporangial formation and larval death were usually simultaneous. A fourth stage larva usually contain about 20,000 sporangia. One, sometimes two, exit tubes were formed by each sporangium and grew out the body wall of the dead larva. A vesicle was formed at the end of each exit tube and the entire contents of the sporangium flowed into the vesicle. Within the vesicle were formed from 4 to 50 zoospores (average of 12/vesicle). The wall of the vesicle disintegrated and the motile zoospores were released into the water to repeat the cycle.

#### c. Sexual reproduction.

In the sexual phase, the individual segments of the hyphae became either antheridia or oogonia. The antheridial protoplast moved through a conjugal pore into an adjacent oogonium to form a zygote. This became the oospore (resting spore) by the formation of a thickened wall around the combined protoplasts. The oospore remained dormant until stimulated to germinate and released motile zoospores into the water to repeat the cycle. Both asexual and sexual cycles occurred within a single larva and also within a single hypha. The cycle most frequently observed was the asexual one. The factors which stimulate the fungus to enter the sexual phase are presently under investigation.

#### 2. Effects on nontarget species.

Although available literature on the Lagenidiales and the genus *Lagenidium* reveals these aquatic fungi to be very host specific, the possible effect of this organism on nontarget aquatic organisms which may be essential food chain species in the environment was considered early in the study. *Cyclops*, *Daphnia*, *Scapholeberis*, several additional unidentified species of copepods and cladocerans, polychaetes, dytiscids and chironomids were collected from naturally occurring sites and tested by placing approximately equal numbers in test and control containers; *Lagenidium* was added to the test containers daily. Mosquito larvae and equal quantities of brewers' yeast and algae as food for the nontarget organisms were periodically added to all containers. The mosquito larvae in all inoculated containers became infected and died. Population estimates and actual counts of the nontarget organisms revealed no greater survival among the control than in the test organisms. Microscopic examination of young and adult organisms revealed no fungal infections.

Adults were observed daily with eggs; abundant young specimens, from newly hatched through all stages, were routinely observed. Microscopic examination of centrifuged detritus revealed no fungal infection of any organism except mosquito larvae.

## B. Field Observations.

### 1. Colusa site.

The data presented in Table 1 show the number of living larvae collected after treatment from sites 1, 2 and 3; the number that were infected; and the percent of infection. It appears from these data that the environmental factors mentioned earlier did have an adverse effect on the fungus at sites nos. 1 and 3. It is also apparent that at site no. 2, the only site with water qualities like those of the rice fields and drainage ditches, that all larvae became infected and all larvae died.

The data presented in Table 2 show the mean daily collections of living larvae and pupae from site no. 2 prior to and after treatment. Although the raw data are recorded for each separate plot and instar, for purposes of this presentation all instars and plots have been combined. Note that the field populations in the treated plots were essentially reduced to zero by the 4th day. All of the larvae collected the 2nd, 3rd and 4th post treatment day were held in the laboratory. All larvae from treated plots were infected and all died within four days. The sites were examined again on the 17th post treatment day. Sites nos. 1 and 3 were totally dry. At site no. 2 living larvae or pupae could be found in the treated plots though the control plots at this site continued to be active producers.

### 2. Hanford site.

On the day of treatment the larvae were primarily second stage *A. nigromaculis* with about 2% of the population early 3rd instar. Of the 100 larvae collected just prior to treatment and set in the laboratory with inoculum, all were dead within two days and all were positive for *Lagenidium*.

On the 2nd post treatment day, the larvae were primarily 4th stage and several dead *A. nigromaculis* larvae were removed from each of the sampling plots in the treated check for microscopic examination. These data revealed that infection had occurred in the natural population, for all of the dead larvae collected were infected.

Also on the 2nd post treatment day, 10 living *A. nigromaculis* larvae were collected from each of the treated plots and held in distilled water for observation. All of the larvae died, but positive identification of the fungus could be made in only 83% of the cadavers. The remaining 17% had all of the characteristics of infection but positive identification of fungal hyphae could not be made due to intense pigmentation of the larvae.

On the 3rd and 4th posttreatment days the population was essentially pupae and the data from the collections on these days (Table 3) suggest that the larvae were either unable to survive confinement in the 4-foot square sampling plots or that they were not sealed as tightly to prevent ingress or egress as we thought. Nevertheless, the three larvae that were collected from the treated plots on the 3rd posttreatment day were infected and died within two days, and no living larvae or pupae were collected with the five standard dips on the 4th posttreatment day. Examination of the entire treated check on the 4th posttreatment day revealed that the water was rapidly receding from the upper, untreated end and was moving the pupae and late 4th instars into the lower, treated end. Numerous living pupae and a few larvae of *A. nigromaculis* were observed at several locations in addition to the large numbers of dead larvae that were generally distributed throughout the lower end of the check.

A diligent search of the three sampling plots in the treated check on the 4th posttreatment day revealed a few living 4th stage larvae and pupae in sampling plots nos. 2 and 3. These were collected and returned to the laboratory for observation. Emergence and survival were normal. Whether these were specimens present when treatment was applied and did not become infected or whether they were specimens that had moved down from the upper, untreated end and worked their way into the sampling plots is unknown. Previous laboratory data (McCray et al. 1973) had shown that pupae and larvae that were late 4th stage when exposed usually did not become infected.

On the 8th posttreatment day, *C. tarsalis* had appeared in that portion of the treated check that still contained water. Numerous *C. tarsalis* larvae were collected from the treated and untreated checks and held in the laboratory for observation. Within three days all from the treated check were dead and all were infected. None from the untreated check died. These data revealed that the fungus had cycled at least one time and infected the newly hatched *C. tarsalis*.

During these studies more than 1400 aquatic organisms from the treated sites were examined. No infections were observed in any of these specimens.

## Discussion

The studies of the life cycle of this strain of *L. gigarteum* and its host-parasite relationships with culicine larvae at TDL revealed two distinct modes of action. These are directly correlated with the asexual and sexual cycles of the fungus.

One mode of action, that involving the asexual cycle, occurs in a permanent (or semipermanent) body of water in which there is continuous oviposition throughout the mosquito breeding season. In such a situation the fungus devel-

ops within the body of the infected larvae and the resulting sporangia produce and liberate into the water, motile, infective zoospores. These zoospores, in turn, infect other larvae present and the cycle is repeated. The time elapsed between infection and subsequent release of zoospores depends upon the stage in which the larvae were infected (McCray et al. 1973) and the temperature of the water (McCray et al. In manuscript). In younger larvae and in higher temperatures the cycle is shorter than in older larvae and lower temperatures. The motile zoospores are short lived and data available at this time indicate that the zoospores probably do not remain infective for more than 48 hours. If there are no larvae present for them to infect, the cycle is interrupted.

The other mode of action, that involving the sexual cycle, occurs in bodies of water that are intermittently dry and flooded. In this situation, the fungus develops within the body of the infected larvae and those hyphal segments that form antheridia and oogonia conjugate to form oospores (resting spores).

Data presently available reveal that these oospores remain dormant within the infected cadavers and are released at the time the cadavers disintegrate. The oospores remain viable after the water has evaporated and data from tests conducted for one year in the laboratory show that the oospores are capable of surviving in the dormant state for that period of time. When the dry dormant spores are flooded, they germinate. Although actual germination has not yet been observed, available laboratory data indicate that motile, infective, zoospores, rather than hyphae, are produced at the time of germination. These zoospores infect the newly hatched larvae and the asexual cycle is resumed.

These two modes of action are, potentially, overlapping cycles, for if a body of water only partially dries up and is then reflooded, those oospores near the edge that were exposed to drying, germinate and contribute to the existing asexual cycle.

In these studies two distinct habitats were utilized. At the Hanford site, an intermittently dry and flooded habitat in which *A. nigromaculis* was the principal species present, this strain of *L. giganteum* did infect natural populations of *A. nigromaculis* and all infected specimens died. Field populations were dramatically reduced within three days after treatment and the *C. tarsalis* which subsequently appeared in the treated pasture became infected and all collected larvae were infected and died.

At the Colusa site, studies of the environment revealed the principal breeding habitat for *C. tarsalis* to be the rice fields and associated drainage ditches. In the single test site which most nearly resembled the normal breeding habitat, a single introduction of *Lagenidium* did infect and eliminate the natural population of *C. tarsalis*. When the test was terminated 17 days after *Lagenidium* was introduced, no larvae could be found in this site. The effect of the fungus was demonstrably reduced in the seepage ditches in which water analysis had revealed conditions known to be detrimental to the fungus. Throughout the studies no nontarget organisms were found to be infected.

These studies revealed that this strain of *L. giganteum* is an excellent candidate for further evaluation and more definitive tests are planned.

#### Acknowledgments

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# HARVESTING EGGS FROM WILD NOTONECTID POPULATIONS

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Over the last several years there has been an increasing interest in the potential use of notonectids for mosquito control. Four species of backswimmers, namely, *Notonecta undulata*, *N. unifasciata*, *N. shooterii* and *N. kirbyi* are reported to be widespread in California (Usinger 1956). Several species have been reported to be efficient mosquito larval predators under laboratory conditions and in limited field trials, (Hinman 1934, Bay 1967, and Lee 1967). Recently there have been some studies on predator prey preference and bioenergetics which add further support to the contention that backswimmers may have a practical role in mosquito control programs. (Ellis and Borden 1970, Toth and Chew 1972).

It is generally acknowledged that where notonectids are abundant larval mosquitoes are not found in great numbers, however, there is little scientific evidence supporting a causal relationship. Notonectids are well known general predators and observations indicate that their prey consists of various aquatic animals including small fish (Usinger 1956). They will also feed on non-aquatic organisms dropped into the water in the laboratory (Bueno 1905, Ellis and Borden 1970).

Characteristically notonectids are found more frequently in permanent to semipermanent waters whereas mosquito problems are more likely to be associated with more transient water sources. Thus a crucial point in a research program is to establish a system for bridging this gap so that the predator coexists in time and place with the mosquito populations. In order to evaluate notonectids as a practical means of mosquito control research is needed in the following areas: (1) a method for production of the predator; (2) a system for accumulation and storage; (3) a technique for release; and (4) survivorship of the predator and evaluation of its impact on mosquito populations. Discussion will be devoted to the first of these research needs.

There are generally two approaches which appear to be promising for fulfillment of the first of the above requirements. One would be to colonize the predator so that the desirable stage of the predator would be available when needed and the second would be to use natural populations as the colony and periodically harvest the desired stage. Both approaches are being taken in this laboratory, however, my remarks will be restricted to the second approach.

All stages of *N. kirbyi* can be collected in the field. Due to the fragile nature of the nymphal instars and the mobility of the adults, these stages were excluded from consideration. The egg stage was considered to be the best candidate because it is relatively easy to handle.

Either colonization of the insect for egg production or harvesting the eggs directly from the field require a system for slowing or halting development so that relatively large numbers can be accumulated for release during the mosquito season. Sjogren (personal communication) has demon-

strated egg storage is practical with *N. unifasciata*. Low temperature storage of *N. kirbyi* appears feasible also.

As mentioned above one approach to utilizing notonectids for mosquito control is to take advantage of naturally occurring populations as a means of supplying individuals for later release. The remainder of this paper will deal with harvesting of eggs from wild *N. kirbyi* populations. This species as well as several others readily use submerged vegetation as oviposition sites in natural water sources. Large numbers of eggs have been observed on living and dead plant material hanging into and emerging from the water sources. In the laboratory this species will lay eggs on a variety of substrates including submerged  $\frac{1}{4}$  inch and  $\frac{1}{2}$  inch hardware mesh cloth, plastic and metal screens, and the walls of glass, metal and wood water containers. It appears that a major requirement is a surface structure that adult females can grasp.

The use of artificial oviposition sites was first attempted at an oxidation pond outflow near Vacaville, Solano County. Broom straws approximately 6 inches, 12 inches and 15 inches long were inserted into 3 ft wooden lath floats. The straws in clusters of three were placed in drilled holes approximately one inch apart so that they descended vertically into the water. Broom straws were selected because they are inexpensive, easy to handle and biodegradable. Eggs were deposited on the straws and counts of up to 2 to 3 eggs per straw for each day of exposure have been recorded. Oviposition has been recorded from December through May, however, it is suspected that the period of oviposition is actually longer than so far observed. The highest levels of egg deposition have been recorded in February, March and April.

The use of floats for securing the broom straws has been abandoned primarily because of vandalism at the study site. An alternative system employing a one inch wide aluminum lath about 4 ft in length with approximately the last 12 inches at each end bent at right angles to form legs has proven satisfactory. Broom straws (3 to 6 per cluster) are secured vertically by inserting the ends through small holes drilled one inch apart through the horizontal portion of the lath. This aluminum rack is then entirely submerged in the proper depth of water so that the legs stick into the mud bottom supporting the horizontal portion two or three inches below the water surface.

A number of aquatic insects use the straws for oviposition sites. These include, besides *N. kirbyi*, *N. undulata*, *N. unifasciata*, *N. shooterii*, several corixid species and a single tropisternus sp.

In summary, the method described above represents only a portion of the first phase of the experimental work necessary in evaluating notonectids as mosquito control agents. It does, however, lend encouragement for continued effort along these lines of investigation.

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## INSECT DEVELOPMENTAL INHIBITORS: EFFECTS ON NON-TARGET AQUATIC ORGANISMS

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### ABSTRACT

Acute, short-term toxicity of ZR-515 (isopropyl 11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate) on various aquatic organisms was studied. In the laboratory, 35 organisms including Protozoa, Platyhelminthes, Rotatoria, Annelida, Arthropoda, Mollusca, chordata, and Thallophyta were tested with technical AR-515. Dosages used for mosquito larvae control created

no adverse effect on most of the organisms tested. In the field (artificial containers, ponds, and irrigated pastures), a slow-release formulation was used with no visible effect on most of the non-target organisms. However, larvae of aquatic Diptera (Chironomidae, Ephydriidae, Psychodidae) showed some sensitivity.

## USE OF DICHLORVOS AGAINST ORGANOPHOSPHATE RESISTANT ADULT MOSQUITO POPULATIONS IN KERN COUNTY

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### ABSTRACT

Application of 0.1 lb/acre dichlorvos (DDVP) in total volume of 6 fluid ounces to 4 gallons applied by air, achieved good control of organophosphate resistant *Aedes nigromaculis* adults under field conditions. Best results were obtained by block or band treatments made parallel to air movement within 90 minutes of sunrise, with < 2 mph wind,

and temperatures between 60 and 85°F. Equivalent results were achieved with a standard ULV system, a high volume spray system or an experimental 450 PSI adulticiding system when petroleum oils with initial distillation boiling point between 420 and 530°F were used as carrier.

# INOCULATION OF HYDRA (COELENTERATA) AND PREDATION EFFECTIVENESS IN EXPERIMENTAL MOSQUITO (*CULEX*) BREEDING HABITATS

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**INTRODUCTION.**—A critical need exists for effective biological control agents in mosquito control as alternatives to chemical larvicides. One potentially useful but little explored group of biological control organisms which attack mosquito larvae are freshwater hydra. Hydra were known to be effective in killing cultured mosquitoes through stinging or consumption ever since Hargreaves (1924) reported a complete engulfment of *Theobaldia* larvae by *Hydra* species in Italy. Additional laboratory studies on *Hydra vulgaris* Pallas have shown this species to be an effective mosquito destroyer (Hamlyn-Harris 1929, Twinn 1931, Stephancides 1960). In 1969 Qureshi and Bay quantified the destructive capacity of *H. americana* Hymen in 1.2 - 12 liter filter aquaria (Bay 1967). Their work showed highly significant reductions in mosquito densities. They suggested that hydra might be used practically if the mass production of thecated embryos were economically feasible. The role of hydra in natural control (Hamlyn-Harris 1929) and their practical use in mosquito abatement (Laird 1956, Stephanides 1960, Qureshi and Bay 1969, Legner and Medved 1972) have been alluded to frequently. Thus, larger scale field experiments were called for as well as an investigation into the comparative behavior of different hydra species. This report discusses results of predation experiments we conducted with the European green hydra, *Chlorohydra viridissima* (Pallas), in simulated natural habitats of *Culex peus* Speiser and *C. pipiens quinquefasciatus* Say during 1972 and elaborates on the characteristics of hydra that indicate their potential effectiveness for biological control.

**MATERIALS AND METHODS.**—Stock cultures of the European strain of *Chlorohydra viridissima*, obtained from the University of California, Irvine<sup>1</sup>, were maintained under controlled laboratory conditions as previously described (Loomis and Lenhoff 1956, Lenhoff and Brown 1970).

Hydra were tested for their adaptability to water in 80 mm petri dishes each filled to a depth of 20 mm with the water to be tested. Twenty *C. viridissima* were evenly-spaced in a given dish and graded for adaptability, according to 3 criteria selected to describe their physical condition.

The gradings were as follows:

Well-adapted = tentacles well stretched and body normal.

Intermediate = tentacles and body half retracted or shrunk.

Poorly-adapted = tentacles and body shrunk into a ball shape, and with rare recovery capability.

In predation tests on *Culex* species, hydra were introduced into 24 separate m<sup>2</sup> fiberglass tanks, each divided in half by 11.8 mesh/cm saran screen with mosquito emergence traps fixed in covers at the top (Figs. 1 and 2). The base of each tank was covered with 3.8 cm of coarse silica sand, into which were plunged 20, 25-cm long bamboo sticks, spaced 10.2 cm apart. Water flow was adjusted at a constant rate of 2600 liters per day for all units. A metal ion neutralizing agent, EDTA versine (Lenhoff and Brown 1970), was released by droplets from above the filtering tank to simulate natural water conditions (Fig. 1). Mosquito larval breeding was stimulated by the introduction of freshly hatched *Culex peus* larvae in 4 density groups of 6 replicates each (25, 50, 100 and 250 larvae per day), beginning one month in advance of hydra introduction. Sufficient larval food was supplied through the addition of 2 g/day of poultry chick starter mash.

Hydra were inoculated in each half of a divided unit at the rate of 500 each applied homogeneously to the whole surface area, using the chilled volumetric method (Lenhoff and Loomis 1957). A large number of *C. peus* larvae, 4,700/day, were easily obtained in August and September in the form of egg rafts from enclosed, controlled, semi-natural breeding systems developed at the Aquatic Research Laboratory Riverside (Legner and Medved 1972—Fig. 1). *Culex pipiens quinquefasciatus* became similarly available in October. Mosquito destruction was assessed through adult mosquito emergence and % destruction calculated with the formula:

$$\% \text{ Destruction} = 100 - \left[ \frac{\text{No. adults emerged with hydra}}{\text{No. adults emerged in control}} \right] \times 100$$

Although a general method of sampling hydra was known for field situations (Kanaev 1952, Southwood 1971), the specific technique developed for our experiment was a random method in which 4 bamboo sticks were drawn at random and attached hydra counted at weekly intervals during the test period (Fig. 3). The adaptability of the population was periodically checked and compared to that of natural pond waters in the vicinity of Riverside (Lenhoff et al. 1969). Growth rates of hydra in the subsequent intervals of time were calculated from the number of hydra counted.

**RESULTS AND DISCUSSION.**—Adaptability of *C. viridissima* to Experimental Water.—Adaptability tests of *C. viridissima* to experimental water showed 91.65 - 93.35% were well adapted in September and October, respectively (Table 1). The experimental water in these tests was similar to natural waters where *Culex* species were found breeding such as Lake Hole (91.65%) and standing water on the University of California, Riverside, campus (90.00%). However

<sup>1</sup>Dr. H. M. Lenhoff, Pathobiology Laboratory.

Table 1.--Proportion of well-adapted<sup>1</sup> *Chlorohydra viridissima* to experimental mosquito breeding water compared with natural waters tested in the laboratory. Riverside, California 1972.

Collection Date	Locality	Mean No. Well-adapted <sup>2</sup>	Percent Well-adapted	Mosquito Breeding
July 8	Lake Evans, Riverside	19.67	98.35	
July 9				
July 8	Lake Evans, Riverside	19.67	98.35	<i>Culex</i> sp.
July 23	Lake Hole, Riverside	18.33	91.65	<i>Culex</i> sp.
Aug. 5	Standing water, UCR campus	18.00	90.00	<i>Culex</i> sp.
Aug. 7	Standing water, UCR housing area	17.33	86.66	<i>Culex</i> sp.
Sept. 1	Experimental water, UCR	18.33	91.65	<i>Culex</i> sp.
Sept. 2	Mockingbird Reservoir, Riverside	18.67	93.35	--
Oct. 1	Experimental water, UCR	18.67	93.35	<i>Culex</i> sp.
Oct. 9	Lake Hole, Riverside	18.67	93.35	-- <sup>3</sup>

<sup>1</sup>Tentacles well stretched and body normal.

<sup>2</sup>20 hydra per each of 3 replicates.

<sup>3</sup>Larval breeding not detected but a biting collection of *Culex pipiens quinquefasciatus* was made.

it was more favorable than putrifying standing water sampled at the university housing area where only 86.66% of the test hydra were well adapted.

**Predation in Experimental *Culex* Larval Habitats.**—The first significant *C. peus* population reduction was 55.46% over all densities recorded one week after hydra inoculation, with the higher percent reductions proportional to host density (Table 2). The predator maintained its effectiveness above the level of 50% destruction throughout the rest of the month in all groups of host densities except the lowest density in which predation appeared unstable, fluctuating between 18.36% and 42%, with an average percent destruction of only 11.95% (Table 2). The average for September showed that the most effective predation occurred at the highest mosquito density (55.02%), the predator acting in an obvious density dependent manner (Table 2). Predation more than tripled when the host density was doubled from the lowest density (25 larvae/day) to the next highest density (50 larvae/day). Predation was enhanced when the area of contact in a given volume of environment was narrowed.

Destruction of *C. p. quinquefasciatus* was greater in October (Table 3), hydra predation being effective in keeping this species at a lower level than *C. peus* in September. Apparently, the hydra population was well established by the end of September (Table 4), and the effect of predation was increased through the growth and multiplication of the hydra. Although a rapid growth rate of green hydra of 1.2 to 3.5 days was reported (Lenhoff and Brown 1970, Muscatine 1961) in our experiment sampling showed a population doubling approximately every 30 days (Table 4). This growth rate was linearly related in time (Slobodkin 1966) in all density groups except the 100 larvae/day density in which the hydra population decreased in the samples taken both on September 16 and 20, for unknown reasons. The

drop in hydra density at the cited host density was accompanied by a slightly lower destructive capacity (Table 3). The hydra population reached its greatest density in the 50 larvae/day host density.

**Attributes of Hydra For Biological Mosquito Control.**—The foregoing results show that hydra are potentially effective as biological control agents for sustained freshwater mosquito larval control. Unlike predacious insects and mosquito fish (*Gambusia*), hydra can be easily mass produced by the millions (Loomis and Lenhoff 1956, Lenhoff and Brown 1970) and no danger of cannibalism exists when they can double in 1.2 - 4 days depending on the species (Lenhoff and Brown 1970). Hydra produce semi-dormant, encysted embryos which may be stored and disseminated with convenience. Because hydra cannot disperse readily beyond their immediate habitat, pending favorable conditions to their survival, they reproduce in the environment into which they are introduced. Hydra, like pathogens, can be used in waters that are too shallow or too temporary for applying larvivorous fishes. Also, hydra kill organisms larger than themselves and they are not host specific. Thus, three basic requirements of biological control organisms are met in hydra, (1) amenability to mass production, (2) ease of storage, and (3) practicality of applying to infested areas.

**Other Advantages.**—Once a hydra has ingested a mosquito larva it initiates a bud giving rise to a new individual. Therefore, the more mosquitoes consumed the greater the chance for increasing in sufficient numbers to eradicate mosquitoes in a habitat.

In the absence of food, hydra do not die but merely reduce their size by losing cells at the tips of their extremities. Growth resumes upon gaining new food and reproduction follows. Green hydra which harbor symbiotic algae in their bodies survive intact longer than those without such sym-

Table 2. Average number adult mosquitoes emerged and percent reduction by *Cylindrohydra viridissima* inoculated in breeding habitats with 4 different densities of *Culex peus* larvae - 6 replicates, September 1 through 30, 1972.

Collection Date		25 larvae/day <sup>1</sup>		50 larvae/day		100 larvae/day		250 larvae/day		Subtotal	
		Con.	Trt.	Con.	Trt.	Con.	Trt.	Con.	Trt.	Con.	Trt.
Sept. 1	Mean No.	11.83	12.50	15.00	11.67	30.17	32.33	35.83	29.17	92.83	83.67
	% Reduct.		-5.63		22.22		-6.62		18.60		7.71
Sept. 5	Mean No.	14.58	15.67	11.50	10.50	24.00	27.58	44.58	35.33	94.67	89.08
	% Reduct.		-7.42		8.69		-14.93		20.74		5.89
Sept. 7	Mean No. <sup>2</sup>	12.33	9.00	14.67	11.50	22.33	8.17	36.00	9.33	85.33	38.00
	% Reduct.		27.02*		21.59		63.43*		74.07*		55.46*
Sept. 9	Mean No.	8.17	9.67	8.33	4.67	11.00	5.50	9.33	2.50	36.83	22.33
	% Reduct.		-18.36		44.00		50.00*		73.21*		39.36*
Sept. 11	Mean No.	7.50	7.33	8.17	3.83	13.83	6.17	11.67	3.50	41.17	20.83
	% Reduct.		2.22		53.06*		55.42*		70.00*		49.39*
Sept. 13	Mean No.	9.50	7.33	6.33	1.17	9.17	6.83	6.50	4.83	31.50	20.17
	% Reduct.		22.80		81.57**		25.45		25.64		35.96**
Sept. 15	Mean No.	13.17	10.67	5.67	2.17	15.83	5.83	10.83	4.83	45.50	21.83
	% Reduct.		18.98*		61.76**		63.15*		55.38*		52.01***
Sept. 17	Mean No.	12.50	10.50	4.33	1.67	21.67	6.17	9.83	3.50	48.33	21.83
	% Reduct.		16.00		61.53*		71.53*		64.40*		54.82**
Sept. 19	Mean No.	10.17	9.17	3.17	2.50	21.00	7.67	12.83	3.17	47.17	22.50
	% Reduct.		9.83		21.05		63.49*		75.32*		52.30*
Sept. 21	Mean No.	9.33	5.33	9.33	8.17	33.00	10.83	14.83	5.33	66.50	29.67
	% Reduct.		42.85*		12.50		67.17*		64.04*		55.39**
Sept. 23	Mean No.	8.83	6.00	4.67	3.83	27.83	5.83	8.83	5.17	50.17	20.83
	% Reduct.		32.07*		17.85		79.04**		41.50		58.47**
Sept. 27	Mean No.	5.00	4.75	5.67	2.67	16.75	5.25	7.58	2.17	34.92	14.83
	% Reduct.		5.00		52.41*		68.31**		70.65*		57.52**
Sept. 29	Mean No.	5.00	4.50	11.33	2.50	22.67	8.00	7.83	3.00	46.83	18.00
	% Reduct.		10.00		77.94*		64.70*		61.70*		61.57***
TOTAL	Mean No.	9.84	8.65	8.32	5.14	20.71	10.47	16.65	8.60	55.52	32.74
	% Reduct.		11.95**		41.24***		50.01***		55.02***		45.07***

<sup>1</sup>Con. = control; Trt. = treatment (Hydra).

<sup>2</sup>% Reduction = 100 - (treatment/control) x 100.

\* = 90% significance; \*\* = 95% significance; \*\*\* = 99% significance (Match-paired "t" test).

bionts (Muscatine and Lenhoff 1965, Slobodkin 1966). Persistence at a size suitable for mosquito predation can be as long as 2 months.

Hydra have evolved a mechanism to form semi-dormant thecated embryos when unfavorable conditions arise such as the drying-up of the habitat, lack of food, or cold (Rutherford et al. 1964), and accumulated metabolites

(Loomis 1964) and these embryos have been observed to remain viable for periods exceeding 2 months.

Hydra can reproduce in a wide range of environmental conditions such as in water pH of 4.2 - 7.8 and in low oxygen tensions (Lenhoff and Loomis 1957, Loomis 1954). Their tolerance of chemical pollution is indicated by their survival for days in 10<sup>-4</sup>M KCN (H. M. Lenhoff, personal



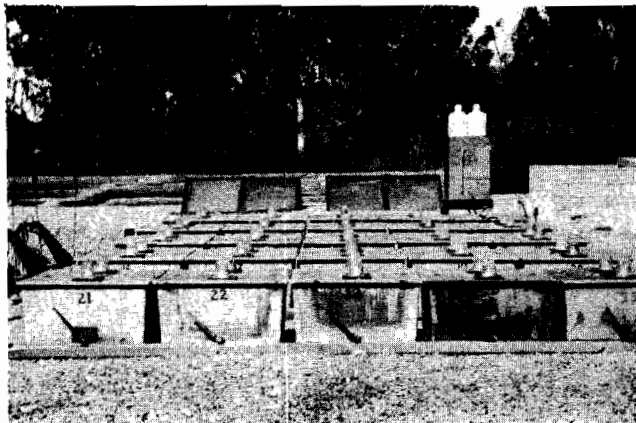


FIG. 1.—One meter-square fiberglass tanks equipped with water flow and quality control and screened plastic emergence traps.

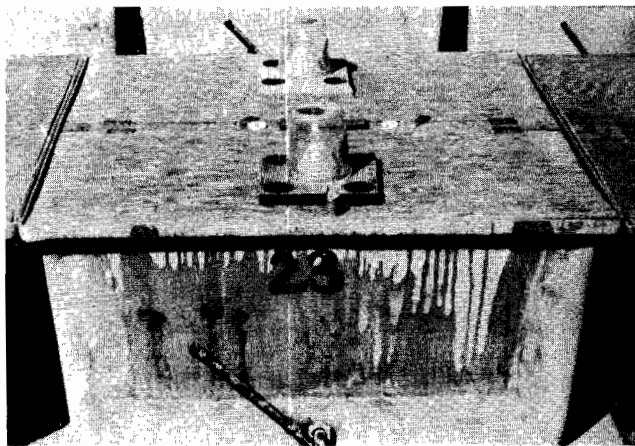


FIG. 2.—An individual one meter-square fiberglass tank showing divided halves and separate emergence traps for treatment and check portions of hydra predation experiments.



FIG. 3.—Hydra sampling materials showing a bamboo stick sample drawn from the treated half of a tank on which hydra are attached. Instantaneous counting is essential for maximum accuracy.

communication). Also, species differ in their tolerance of temperatures with observed ranges of 38 - 40°C. (Eakin 1961, Loomis 1954). Therefore, it may be best to use hydra species that are naturally present and adapted to particular problem habitats for mosquito control.

Continued studies in conjunction with Dr. H. M. Lenhoff of the University of California, Irvine, will explore the possibilities for using hydra as biological control agents of mosquito larvae. Sufficient quantities of selected hydra species will be produced for large scale field trials against *Culex tarsalis* and *Aedes nigromaculis* in California. Determinations will be made of the most effective species of hydra for different seasons and different conditions of water quality; and methods sought for mass production, retrieval and storage of hydra cysts as well as for breaking cyst dormancy.

#### Acknowledgment

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Table 3.—Average number adult mosquitoes emerged and percent reduction by *Chlorohydra viridissima* inoculated in breeding habitats with 4 different densities of *Culex pipiens quinquefasciatus* larvae -- 6 replicates, October 1 through 31, 1972.

Collection Date		25 larvae/day <sup>1</sup>		50 larvae/day		100 larvae/day		250 larvae/day		Subtotal	
		Con.	Trt.	Con.	Trt.	Con.	Trt.	Con.	Trt.	Con.	Trt.
Oct. 1	Mean No. <sup>2</sup>	9.67	4.00	4.00	2.67	24.50	6.50	7.50	0.50	46.67	13.67
	% Reduct.		58.62*		46.67*		73.47***		93.33***		70.71***
Oct. 3	Mean No.	4.50	2.17	4.00	0.50	16.00	8.83	6.83	1.67	31.33	10.17
	% Reduct.		51.85*		87.50**		63.54**		75.60**		67.55***
Oct. 5	Mean No.	7.00	3.83	2.50	1.00	19.83	11.00	11.50	3.33	40.83	19.17
	% Reduct.		45.23*		60.00		44.54*		71.01***		53.06***
Oct. 7	Mean No.	6.33	4.17	4.33	1.67	19.00	8.67	8.67	0.33	38.33	14.50
	% Reduct.		34.21*		61.53*		56.14**		96.15*		62.17**
Oct. 9	Mean No.	8.50	4.33	1.50	0.33	17.00	5.83	5.17	1.00	32.17	11.50
	% Reduct.		49.01**		77.77**		65.68**		50.65***		64.24***
Oct. 11	Mean No.	4.00	0.33	2.17	0.50	5.67	1.67	5.00	1.50	16.83	4.00
	% Reduct.		91.66**		76.92*		70.58*		70.00*		76.23***
Oct. 13	Mean No.	1.00	0.17	0.17	0.00	2.50	0.67	0.67	0.33	4.33	1.17
	% Reduct.		83.33**		100.00***		73.33**		50.00		73.07**
Oct. 17	Mean No.	1.17	0.25	0.25	0.17	2.58	0.75	0.83	0.33	4.83	1.50
	% Reduct.		78.57**		33.33		70.96**		60.00**		68.96***
Oct. 21	Mean No.	2.25	1.25	1.75	0.33	9.08	2.75	6.83	0.75	19.92	5.08
	% Reduct.		44.44**		80.95***		69.72***		89.02***		74.47***
Oct. 25	Mean No.	6.25	1.50	0.92	0.00	7.08	4.67	5.75	1.83	20.00	8.00
	% Reduct.		76.00*		100.00*		34.11*		68.11*		60.00**
Oct. 29	Mean No.	2.75	1.67	2.17	0.25	8.25	4.42	7.67	1.25	20.83	7.08
	% Reduct.		57.57*		88.46*		46.46*		83.69**		66.00**
TOTAL	Mean No.	4.86	2.11	2.25	0.67	11.95	4.76	6.04	1.17	25.10	8.71
	% Reduct.		60.95***		73.92***		60.77***		76.14***		66.95***

<sup>1</sup>Con. = control; Trt. = treatment (Hydra).

<sup>2</sup>% Reduction = 100 - (treatment/control) x 100.

\* = 90% significance

\*\* = 95% significance

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Table 4.—Average number of *Chlorohydra viridissima* sampled from 4 randomly selected bamboo sticks per tank replicate following inoculation in *Culex* spp. larval breeding habitat of 4 different densities -- 6 replicates, September 7 through October 31, 1972.

Sample Date	Average Number Hydra per 4 Sticks				Subtotal
	25 larvae/day	50 larvae/day	100 larvae/day	250 larvae/day	
Sept. 8	23.33	74.83	94.67	73.00	265.83
Sept. 12	35.83	78.50	170.17	107.17	391.67
Sept. 16	43.50	101.50	190.50	87.83	423.33
Sept. 20	48.00	114.33	104.83	88.50	355.67
Sept. 24	87.50	162.83	111.50	118.00	479.83
Sept. 28	127.83	198.50	107.67	120.33	554.33
Oct. 8	146.83	210.50	139.50	154.17	651.00
Oct. 16	180.83	203.83	129.83	184.17	698.67
Oct. 24	173.83	225.17	121.33	207.67	728.00
Oct. 31	164.33	229.33	123.00	207.67	724.33

# POND TESTS WITH ZR-515: BIOLOGICAL AND CHEMICAL RESIDUES

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**INTRODUCTION.** In a previous report (Schaefer and Wilder 1972) we demonstrated that the developmental inhibitor ZR-515 (isopropyl 11-methoxy-3, 7, 11-trimethyl-2, 4-dodecadienoate) had the potential of controlling OP-resistant *Aedes nigromaculis* larvae under field conditions, at rates as low as 1/8 lb/A. However, the applications had to be carefully timed so that medium to late 4th stage larvae were treated; the residues following application of ZR-515 EC (emulsifiable concentrate) were not sufficient to control earlier stage larvae. During 1972, further studies showed that when EC applications were very carefully timed (against late 4th stage larvae) doses as low as 1/8 lb/A were highly effective against field populations; it was also quite apparent that such treatments were not effective when mixed or earlier aged populations were the target (Schaefer and Wilder 1973). It was obvious that such critical timing of treatments would not allow for operational feasibility. Recognizing that ZR-515 had high inherent biological activity against *A. nigromaculis* we sought to find formulations that would offer longer residual activity.

**METHODS AND RESULTS.** Numerous formulations were evaluated during 1972 that had no apparent increase in residual biological activity as compared to the EC. However, one formulation was found that showed real promise; this was a 10% flowable liquid, slow-release formulation, which was composed of microencapsulated particles containing ZR-515 in a water base. On July 1, 1972 a pond was treated with this formulation at a dose of 0.35 lb AI/A (AI = active ingredient). The bioassay results are given in Table 1.

Table 1. Bioassay of pond water treated with 0.35 lb AI/A of 10% flowable liquid formulation of ZR-515.

Sampling time (days after treatment):	0 <sup>1</sup>	1	2	3	4	5
Bioassay results (% mortality) <sup>2</sup>	100	97	80	75	17	5

<sup>1</sup>Immediately after treatment.

<sup>2</sup>In percent inhibition of adult emergence of 4th-stage *Culex pipiens quinquefasciatus* larvae added and reared in the laboratory.

Based on these findings, we then conducted extensive tests using this 10% flowable liquid formulation, by hand sprayer and by aircraft, on field populations of OP-resistant *A. nigromaculis* larvae. Tests in both the San Joaquin and Sacramento valleys showed that this formulation could achieve practical control of mixed-stage populations of *A.*

*nigromaculis* and good results were obtained at doses down to 1/40 lb AI/A (Schaefer and Wilder 1973).

Since it appeared that this formulation would provide a practical tool for controlling *A. nigromaculis* populations, cooperative tests were made with the personnel of the U. S. Bureau of Sports Fisheries and Wildlife at the Kern Research Area (20 miles west of Delano, California). Formulations were supplied by Zoecon Corporation. The ponds (30 ft x 60 ft x 1 ft) were treated at a rate which appeared likely for the operational control of mosquito larvae (.05 lb AI/A) and at 5 and 10 times this rate. Two formulations were tested at each rate: (1) the 10% flowable liquid described above, and (2) a 5% flowable liquid similar to the 10% except that the microencapsulated particles contained a 1:1 mixture of ZR-515 and Tenox®, a commercial antioxidant. The test formulations were mixed with approximately 2/3 gallon of water in a hand sprayer and applied to a given test pond. Applications were made at early morning hours before wind drift was significant. Shortly after treatment and at 24-hour intervals thereafter, a water sample was taken along the north and the south borders of the control and treated ponds; these were bioassayed with *Culex pipiens quinquefasciatus* larvae by procedures previously described (Schaefer and Wilder 1972).

The results of bioassaying pond water immediately after treatment and then at daily intervals for 3 days is shown in Table 2; it appears that the Tenox formulation offered more residual activity on the third day at all 3 doses. These results were very encouraging since a high degree of biological activity continued for a 3-day period. Also, the bioassay

Table 2. Bioassay of water from treated ponds with 4th-stage *Culex pipiens quinquefasciatus* larvae (in % inhibition of adult emergence).

Formulation	Dose (lb AI/A)	Sampling time (days after treat.)			
		0	1	2	3
10% Std. <sup>1</sup>	.05	100	49	62	0
5% T. <sup>2</sup>	.05	100	40	40	32
10% Std.	.25	100	82	100	46
5% T.	.25	100	91	100	59
10% Std.	.50	100	86	100	49
5% T.	.50	100	82	100	73

<sup>1</sup>10% flowable liquid composed of microencapsulated particles containing ZR-515 in a water base.

<sup>2</sup>5% flowable liquid composed of microencapsulated particles containing a 1:1 mixture of ZR-515 and Tenox in a water base.

Table 3.—Residues (ppm) of ZR-515 in pond water of the Kern Research Area ponds as determined by gas-liquid chromatography.

Formulation	Dose (lb AI/A)	Sampling time (hours after treatment)									
		2		12		24		48		72	
		north	south	north	south	north	south	north	south	north	south
10% Std. <sup>1</sup>	.05	.076	.046	.010	.023	.007	.006				
5% T. <sup>2</sup>	.05	.056	.075	.014	.017	.008	.008	-	-	-	-
10% Std.	.25	.199	.395	.058	.030	.018	.018	.008	.010	.009	.008
5% T.	.25	.260	.279	.056	.083	.026	.036	.013	.012	.009	.011
10% Std.	.50	.320	.544	.080	.103	.028	.025	.010	.022	.010	.010
5% T.	.50	.204	.296	.084	.190	.052	.034	.025	.018	.011	.009

<sup>1</sup>10% flowable liquid composed of microencapsulated particles containing ZR-515 in a water base.

<sup>2</sup>5% flowable liquid composed of microencapsulated particles containing a 1:1 mixture of ZR-515 and Tenox in a water base.

was done using *C. p. quinquefasciatus* larvae, which are much more tolerant to ZR-515 than are *A. nigromaculis*.

For chemical analysis, 600 ml water samples were taken from the upper 2 inches, along the north and south borders of the control and treated ponds; these were extracted in a field laboratory adjacent to the ponds and the extracts were transported to the Fresno Laboratory for quantitative analyses by gas-liquid chromatography. Details of this procedure have been described (Schaefer and Dupras 1973).

The chemical residues are shown in Table 3. At 2 and 12 hours following treatment, 5 of the 6 treated ponds showed higher amounts of ZR-515 along the south border, which was the lee side; but this difference does not appear to exist at 48 or 72 hours. The chemical data does not show a greater concentration of ZR-515 in the 5% Tenox treated ponds (as compared to the 10% standard formulation) at 72 hours, or 3 days, as indicated by the bioassay data (Table 2). Further comparisons of these formulations seem necessary.

The chemical and biological assays of the treated ponds show a steady and rather rapid depletion of ZR-515 from the water; this indicates that ZR-515 dissipates quickly and

is a favorable attribute in relation to current concerns relative to environmental pollution resulting from pesticide use. Other chemical residue studies conducted during 1972 have given similar results (Schaefer and Dupras 1973).

#### Acknowledgment

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# LABORATORY AND EXPERIMENTAL AND OPERATIONAL FIELD EVALUATION OF MOSQUITO LARVICIDES

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**INTRODUCTION.**—Studies on the efficacy of new and commercially available insecticides against mosquito larvae were initiated in the laboratory and field. Even though larvae of several species of mosquitoes manifest low to high levels of tolerance to many currently used organophosphate insecticides, there still are good possibilities for finding effective compounds in this group of chemicals which will be effective against many resistant populations for a period of time. With these objectives in mind the current laboratory and field investigations were embarked upon to evaluate various experimentally available compounds. Emphasis was placed on compounds which have good potential for becoming commercial for the control of crop and other insect pests. It is very likely that materials developed for these purposes will be more economical to use in mosquito control than those specifically developed for the abatement of mosquito populations. Several of the compounds studied are of novel structures and have different mode of action than the organophosphate larvicides.

**METHODS AND MATERIALS.**—Biological activity of the various compounds in the laboratory was studied in the manner described by Mulla et al. (1966, 1969, 1970, 1972). Technical materials were dissolved in acetone or other appropriate solvent. Serial dilutions were made from the stock solutions (1.0 or 0.1%) and tested against 4th stage larvae of a laboratory colony of *Culex p. quinquefasciatus*. The tests were conducted in 4-oz treated paper cups, each supplied with 100 ml tap water (pH 7.5-8.0) and 20 4th instar larvae. The toxicants were added to the cups as 1 ml or less of the desired toxicant solution. After treatment the cups were maintained at room temperature (75-78°F). (Controls were run concurrently with each test.) After 24 hours of exposure mortality of larvae was assessed in each cup. Moribund larvae were counted as dead. To obtain a representative LD-P line, each material was tested at 3-5 concentrations on 3-4 different days. Each concentration was replicated 2-3 times on a given day. The values obtained were averaged, and a dosage response line was drawn for each material and the LC<sub>50</sub> and LC<sub>90</sub> determined.

In experimental pond studies, emulsifiable concentrate or other formulations of the various materials were applied as dilute sprays by means of a pressurized can sprayer or household window washer. The total volume of spray applied was 2-8 gallons per acre. For assessing the larval population, 5-10 dips were taken before and 24 or 48 hours post-treatment. The level of control was calculated from the larval counts pre- and post-treatment (24-48 hours).

In field evaluation, emulsifiable concentrate formulations were diluted with water or ARCO larvicidal oil and applied by air or ground equipment. The volume of spray applied by the ground equipment was in the range of 5-10 gallons per acre, while the volume applied by air was from a few ounces to 0.5 gallon per acre. Larval populations were assessed by taking anywhere from 10-30 dips per plot or field pre- and post-treatment (24 hours). The level of control was calculated from the mean number of larvae found in pre- and post-treatment samplings.

A 5% granular formulation of carbofuran (Furadan®) was extensively tested. The material was applied manually by using horn seeder applicators. This method of application does not provide for good penetration of the granules through plant cover. An ideal application method for granules will be broadcasters mounted on airplane, a method by which the granules will probably penetrate much better due to the down draft of the plane.

Chemical description of some of the compounds reported on here are:

- Accothion (Sumithion): 0,0-dimethyl 0-(4-nitro-m-tolyl) phosphorothioate
- Aldrin (photo): hexachlorohexahydro-endo, exo-dimethanonaphthalene
- Allethrin: 2-allyl-4-hydroxy-3-methyl-2-cyclopenten-1-one ester of 2,2-dimethyl-3-(2-methyl-propenyl) -cyclopropanecarboxylic acid
- Bay HOX-1619: 2-chloro-5,5-diethyl-2-thiono-1,3,2-dioxaphosphorinane
- Bay KUE-2302: 2-isopropoxyphenyl N-methyl-N-dichlorofluoromethylmercaptocarbamate
- Bay KUE-2327: trichloro analog of above
- Bay MEB-6046: 3,4-dichloro-2-(trichloromethyl)-benzyl-alcohol-acetate
- Bay SRA-7660: phenylglyoxylonitrile oxime 0,0-dimethyl-phosphorothioate
- Bay ZUM 673: 2-(2-Dioxolanyl)-phenyl N-methyl-N-dichlorofluoromethylmercaptocarbamate
- Bay ZUM-676: 2,Cyclopentylphenyl N-methyl-N-dichlorofluoromethylmercaptocarbamate
- Bay 88941: 2-mthyl phenylglyoxylonitrile oxime 0,0-diethyl phosphorothioate
- Carzol®: 3 - dimethylaminomethyleneiminophenyl -N-methylcarbamate, hydrochloride
- Cidal® (Phenthoate): ethyl mercaptophenylacetate, 0,0-dimethyl phosphorodithioate
- Cygon®: 0,0-dimethyl S-(N-methylcarbamoylmethyl) phosphorodithioate
- Dimethrin: 2,4-dimethylbenzyl 2,2-dimethyl-3-(2-methyl-propenyl) cyclopropanecarboxylate
- Dyfonate®: 0-ethyl S-phenyl ethylphosphonodithioate
- Galecron® (ciba 8514): N<sup>1</sup>-(4-chloro-o-tolyl)-N-N-dimethylformamide

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Gardona®: 2-chloro-1-(2,4,5-trichlorophenyl) vinyl dimethyl phosphate  
 Matacil®: 4-dimethylamino-m-tolylmethylcarbamate  
 Me-Dursban® (M-3196): 0,0-dimethyl 0,3,5,6-trichloro-2-pyridyl phosphorothioate  
 N-2596: O-ethyl-S-(p-chlorophenyl ethane phosphonodithioate)  
 Padan®: 0-(2,5-dichloro-4-bromophenyl)0-methylphenylthionophosphate  
 Pyrethrin: pyrethrum (principally from plant species *Chrysanthemum cinerariaefolium*)  
 SBP-1382: (5-benzyl-3-furyl) methyl 2,2-dimethyl-3-(2-methylpropenyl) cyclopropanecarboxylate  
 Supracide®: S-(2-methoxy-5-oxo-2,1,3,4-thiodiazolin-4-yl)methyl)0,0-dimethyl phosphorodithioate

**RESULTS AND DISCUSSION.** Laboratory.—The biological activity of a number of new experimental and semi-commercial and commercial materials was determined against 4th stage larvae of *C. p. quinquefasciatus* (Table 1). Of 26 materials evaluated, 17 showed high level of activity (LC<sub>90</sub> <0.1 ppm) against the test mosquito. It is worth noting that four materials (Bay MFB-6046, Bay ZUM-676, Bay KUE-2302, and Cidial) were quite fast acting against larvae, producing mortality in 3-4 hrs. Bay ZUM-676 was also an excellent pupicide.

Basic H and Bactozyme were not effective against larvae.

Field.—In pond studies of Midgeville (University of California, Riverside), primiphos-methyl proved very effective against two species of mosquitoes, yielding complete control at 0.05 lb/acre active material (Table 2). Bay HOX-1619 also yielded good control of larvae, producing complete kill at 0.1 lb/acre active material. BASF-2350, a carbamate material, did not produce good control of larvae at the rate of 0.5 lb/acre.

In the Oasis ponds (Coachella Valley, Riverside County, California), most materials evaluated produced excellent control of two species of mosquitoes (Table 3). Bay ZUM-676 was especially effective even at a dosage of 0.01 lb/acre active material.

Actellic (primiphos-methyl), as it did in the Midgeville ponds, proved effective in the Oasis ponds, yielding complete control of larvae at 0.05 lb/acre. Cidial (phenthoate) also was effective yielding complete control at 0.05 lb/acre.

Three experimental Bayer compounds were evaluated, but only two yielded complete control at the rate used. Bay 91273 did not produce complete kill of larvae at the rate used. Bacto-zyme, an organic brew or blend of unknown composition, did not yield larval control at the 1.0 gal/acre rate.

In early 1973 when the water temperature was relatively cool (42-55°F), heavy breeding of *Culiseta inornata* occurred in the Oasis ponds. *Culex tarsalis* was absent from the ponds during this period. Several materials were evaluated at two dosages each. In these tests N-2596 proved highly effective, yielding almost complete control of larvae of this mosquito at 0.01 lb/acre (Table 4). Bay KUE-2302, Bay SRA-7660, Bay ZUM-677 and Bay MFB-6046 also yielded excellent control at dosages ranging 0.01-0.05 lb/acre. Bay ZUM-673 was the least effective compound in the range of dosages applied.

Table 1.—Activity of various mosquito larvicides against 4th stage larvae of *Culex pipiens quinquefasciatus* in the laboratory (1972).

Chemical	Lethal Concentration (ppm)		Remarks
	LC <sub>50</sub>	LC <sub>90</sub>	
Bay ZUM-0673	0.048	0.075	
Bay MFB-6046	0.022	0.050	fast acting
Bay ZUM-0676	0.006	0.011	fast acting on larvae and pupa
Bay KUE-2302	0.005	0.009	effective on pupae
Bay ZUM-0677	0.055	0.088	
Bay KUE-2327	0.044	0.065	
Bay HOX-1619	0.004	0.008	
Bay SRA-7660	0.003	0.005	
N-2596	0.0037	0.007	
Dyfonate	0.088	0.150	
MeDursban (M-3196)	0.001	0.002	
Cidial (Phenthoate)	0.004	0.008	fast acting
Accothion	0.006	0.011	
Photo aldrin	0.0015	0.003	
AC-78373	0.003	0.005	
SBP-1382(C 40%)	0.004	0.006	
Pyrethrin	0.012	0.020	
Allethrin with Pip Butoxide (1:2)	0.020	0.040	
Gardona	0.180	0.320	
Padan	>0.100	-	
Dimethrin with Pip Butoxide (1:1)	>0.100	>0.500	
Galecron (Ciba 8514)	>0.100	-	
Cygon	>0.100	-	
BASF-235 I	>0.100	-	
Bay 88941	>0.100	-	
Curzol	>0.100	-	
Matacil	>0.100	-	

*C. inornata* larvae in the Coachella Valley, unlike larvae of *C. tarsalis*, are quite susceptible to organophosphate insecticides. The former mosquito, being a winter mosquito, is not subjected to operational larvicidal treatments. It is possible that *C. tarsalis* larvae which show some tolerance might require slightly higher dosages of the materials tested.

To determine the efficacy of two of the more effective compounds which are commercially available or close to becoming commercial, Primiphos-Methyl and Cidial were operationally tested by applying the materials with ground equipment and aircraft to various breeding sources of *C. tarsalis* and *Aedes nigromaculis* in Kern County, California.

Table 2.- Evaluation of new mosquito larvicides against larvae of *Culex tarsalis* and *C. peus* in experimental breeding ponds (Midgeville, 1972).

Material and Formula	Dosage lb/A	Avg. no. larvae/ dip		% Control
		Pre	Post 24 hr.	
Primiphos-methyl				
EC <sub>4</sub>	0.01	6	2	67
	0.05	6	0	100
	0.10	14	0	100
Check	-	8	13	-
Bay HOX 1619				
EC <sub>2</sub>	0.01	-	-	58
	0.05	-	-	65
	0.10	-	-	100
BASF 2350 EC <sub>2</sub>	0.10	-	-	16
	0.25	-	-	15
	0.50	-	-	32

Table 3.- Evaluation of mosquito larvicides against larvae of *Culex tarsalis* and *Culiseta inornata* in experimental breeding ponds (Oasis, CA., April 1972).

Material and Formulation	Dosage lb/A	Avg. no. larvae/dip		% Control
		Pre	Post	
Bay ZUM-676				
EC <sub>2</sub>	0.01	4	0.4	91
	0.05	3	0.0	100
	0.10	5	0.0	100
Bay KUE-2302				
EC <sub>2</sub>	0.01	7	6.8	3
	0.05	4	0.0	100
	0.10	5	0.0	100
Check	-	2	3.6	-
Actellic EC <sub>4</sub>	0.01	4.4	4.6	0
	0.05	4.0	0.1	98
	0.10	4.6	0.0	100
Cidial EC <sub>4</sub>	0.01	6.7	4.5	36
	0.05	4.8	0.0	100
	0.10	4.8	0.0	100
Bay 93820*				
EC <sub>2</sub>	0.1	25	0	100
Bay 77049*				
EC <sub>2</sub>	0.1	49	0	100
Bay 91273*				
EC <sub>2</sub>	0.1	37	12	68
Bacto-Zyme	0.5 gal	5.0	3.2	36
	1.0 gal	4.2	8.4	0
Check		19	32	0

\*Toxic to Mayflies at rates used.

Primiphos-methyl applied at the rate of 0.1 lb/acre by ground equipment produced complete control of larvae in all treatments except three (Table 5). In one case control was nil and in the other two cases the level of control was quite high. Cidial produced essentially the same trend, producing incomplete (but still high) control in two applications.

The lack of control with primiphos-methyl and Cidial in the four situations cannot be attributed solely to the existence of resistance to OP compounds in the treated populations. Populations characterized with high level of OP resistance are those at Freeborne and Jessup, and yet, at one of these locations, primiphos-methyl yielded complete control. *A. nigromaculis* in Rancho Santa Maria is characterized with low level of tolerance to OP compounds, and yet this population was not completely controlled by the application of Cidial. It, therefore, seems probable that both resistance and/or problems of application, weather, and penetration were responsible for low level of efficacy.

Table 4.- Effectiveness of various mosquito larvicides against 4th stage larvae of the mosquito *Culiseta inornata* in experimental breeding<sup>a</sup> ponds. (Oasis, CA., Jan. 1973).

Chemical and Formulation	Dosage lb/A	Avg. no. of larvae/dip		% Re-duction
		Pre-treat.	Post-treat.	
<b>Test I 1/8/73</b>				
N-2596-EC <sub>4</sub>	0.01	14	0.15	99
	0.05	11	0	100
Bay KUE -2302				
EC <sub>4</sub>	1.01	6	5	17
	0.05	12	0.5	95
Bay SRA-7660				
EC <sub>2</sub>	0.01	12	6	50
	0.05	15	2	87
Check	-	7	8	0
<b>Test II 1/10/73</b>				
Bay ZUM-0677				
EC <sub>4</sub>	0.025	10	1	90
	0.05	8	0	100
Bay MFB-6046				
75 WP	0.025	15	12	20
	0.050	6	0.3	95
Bay ZUM-0673				
50 WP	0.025	7	5	30
	0.050	9	12	0
Check	-	4	6	0

<sup>a</sup>Chemicals were mixed with 90 ml of water (2 gal of spray/A) and applied with one pint all purpose sprayer. Ten dips were taken prior to treatment and 48 hours after treatment.



Table 5.—Field evaluation of primiphos-methyl (EC<sub>5</sub>) and Cidial (EC<sub>4</sub>) against mosquito larvae in various breeding sources, applied as dilute aqueous sprays by ground equipment at the rate of 0.1 lb/acre active ingredients (Kern County, Calif., 1972)

Date	Location	Habitat	Species <sup>a</sup>	Avg. no. larvae/dip		% Control <sup>b</sup>		
				Pre	Post			
<b>PRIMIPHOS-METHYL</b>								
May	23	Tracy	Slough	Ct	15	0	100	
			Pasture	An	20	0	100	
	24	Maple Leaf	Slough	An	18	5	72	
			SA Camp	Pasture	Ct	5	0	100
		Voth	Alfalfa	Ct	8	0	100	
			Santa Maria	Pasture	Ct	10	0	100
		Bryant	Ditch	An	4	0	100	
			Ditch	Ct	4	0	100	
		25	Bryant	Ditch Ditch	An	40	5	88
				Santa Maria	Pasture 7	Ct	10	0
			Santa Maria	Pasture 5,6	Ct	15	0	100
		30	Jessup	Pasture 6A	An	15	15	0
	Freeborne			Pasture	An(R)	20	0	100
	Freeborne		Ditch	Ct(R)	10	0	100	
	Houchin		Ditch	An	10	0	100	
	Roamini		Ditch	Ct	15	0	100	
	Ghilarduci		Ditch Ditch	Ct	20	0	100	
June	5		Arvin	Orchard	Ct	15	0	100
		Albert Angus	Slough	Ct	300	0	100	
	Baldwin	Water ValesValves	Ct	3	0	100		
June	15	W. Lamont	Reservoir	Ct	7	0	100	
		City Sewer	Ditch	Cq	11	0	100	
	29	Dewyn	Dairy	Cq	20	0	100	
		Houghton	Alfalfa	An	5	0	100	
		E. Union	Alfalfa	Mix	6	0	100	
July	31	Santa Maria	Pasture 10	An	10	1	90	
		Santa Maria	Pasture 74	An	16	2	84	

<sup>a</sup>Ct = *Culex tarsalis*, Cq = *Culex quinquefasciatus*, An = *Aedes nigromaculis*. (R) = highly OP resistant, others are characterized with low to moderate level of resistance.

<sup>b</sup>In most treatments with primiphos-methyl, marked pupal mortality occurred. Pupal mortality also occurred in few treatments of Cidial. Pupal mortality not included in % control.

In aerial field applications Phosvel yielded excellent control of less OP-resistant larvae in Rancho Santa Maria, but did not satisfactorily control highly OP-resistant larvae of Grant Pasture (Table 6). When ARCO larvicidal oil was used as the carrier, level of control was mostly unsatisfactory with the application of Phosvel, Supracide and Cidial. Although direct comparative data for the efficacy of water and oil as carriers or diluents are not at hand, there is an indication that ARCO larvicidal oil, when used in this capacity, does not lead to the best results.

Carbofuran granules (5%) applied operationally by hand equipment, at the rate of 0.2 lb/acre active material to several breeding sources, yielded erratic control of larvae (Table 7). In most cases the larval population was *C. tarsalis* and the water pH high 7.5+. In earlier studies it was found that 5% carbofuran granules did not yield desired control

of *C. tarsalis* in Oasis ponds where water pH is over 8.1 (Mulla et al. 1969). However, this material produced excellent control of moderately to highly OP-resistant *A. nigromaculis* (Sanchez Pasture, Tulare County, and Smith Pastures, Kern County), making pre- and post-hatch applications (Mulla et al. 1970).

The present failure of carbofuran in these studies could be attributed to species specificity and/or pH of the water.

From these studies it appears that some OP compounds are still relatively effective against resistant mosquitoes. These materials, if commercialized for crop insect control, could also be utilized for a period of time in the control of susceptible and resistant mosquito populations under many conditions where severe mosquito problems prevail.

Table 6.—Field evaluation of mosquito larvicides against *Culex tarsalis* and *Aedes nigromaculis* applied at the rate of 0.1 lb/acre active ingredients by aircraft to various mosquito breeding sources (Kern County, Calif.)

Material and Formulation	Location	Habitat <sup>a</sup>	Carrier	Vol/A	Avg. no. larvae/dip		% Control
					Pre	Post	
Phosvel EC <sub>2</sub>	Santa Maria	Pasture	ARCO LO	5 oz	9	0	100
	Grant	Pasture (R)	Water	½ gal	7	3	57
	Universal	Duck Club	ARCO LO	½ gal	11	11	0
Supracide EC <sub>2</sub>	Grant	Pasture (R)	ARCO LO	½ gal	10	10	0
	Grant	Pasture (R)	ARCO LO	½ gal	14	7	50
Cidial EC <sub>4</sub>	Santa Maria	Pasture 71	ARCO LO	6 oz	16	5	69
	Santa Maria	Pasture 7	ARCO LO	6 oz	25	23	8
	Santa Maria	Pasture 74	ARCO LO	6 oz	13	1	95
	Malofy	Desert	ARCO LO	6 oz	2	0	100
	A & B	Duck Club	ARCO LO	6 oz	3	<1	75
Pasuja	Duck Club	ARCO LO	6 oz	4	<1	92	

<sup>a</sup>R = species characterized with high level of OP resistance. The remaining populations possessed low to medium level of tolerance to OP mosquito larvicides.

Table 7.—Field Evaluation of Furadan (5% granules) against mosquito larvae in various breeding sources of OP-resistant mosquitoes (Kern County, Calif., 1972)<sup>a</sup>.

Date	Location and Habitat	pH	Avg. no. larvae/dip		% Control	Spp. <sup>b</sup>	
			pre	post			
August	18	Rim ditch overflow	8	37	39	0	Ct
	23	J. T. Sanders alfalfa	7.5	10	1	90	Ct, An
	25	Rim ditch seepage	9	31	25	19	Ct
	28	Rim ditch					
	28	Rim ditch seepage	9	10	7	30	Ct
	30	Rim ditch seepage	8.5	18	8	56	Ct
September	5	Rim ditch seepage	7	14	.3	98	Ct
	7	Gypsum mine	9	48	37	23	Ct, An
	14	L.A. Athletic Club	9	9	.9	90	Ct
	18	Rim ditch seepage	7.5	18	5	73	Ct
	20	Paloma Slough	10.5	39	38	3	Ct
	26	Rim ditch overflow	7.5	38	27	29	Ct

<sup>a</sup>All applications made by hand to 400-600 ft<sup>2</sup> plots, at the rate of 0.2 lb/acre active ingredient.

<sup>b</sup>Ct = *Culex tarsalis*, An = *Aedes nigromaculis*.

#### Acknowledgment

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# EVALUATION OF PROMISING PETROLEUM OILS FOR THE CONTROL OF ORGANOPHOSPHORUS RESISTANT *Aedes nigromaculis* MOSQUITO LARVAE IN IRRIGATED PASTURES

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**INTRODUCTION.**—In 1971 the outlook for mosquito suppression in irrigated pastures of the San Joaquin and Sacramento valleys of California appeared unfavorable owing to resistance to all available organophosphorus larvicides (Womeldorf et al. 1972). Several petroleum oils also failed to achieve satisfactory control when applied by air at the rate of 2.0 gallons per acre. Results were either inconsistent or completely unsatisfactory against the larval stages of *Aedes nigromaculis* in irrigated pastures (Darwazeh et al. 1972, Darwazeh and Ramke 1972, Schaefer and Ramke 1971, and Gillies et al. 1971).

In response to requests from several mosquito abatement agencies and with the cooperation of oil companies, the studies reported upon here were undertaken to evaluate newly developed petroleum oils under laboratory and field conditions to determine their possible use as mosquito larvicides in the multiresistant areas.

**METHODS AND MATERIALS.**—Fifteen petroleum oils were evaluated in the laboratory against fourth instar larvae obtained from a laboratory colony of *Culex pipiens quinquefasciatus* known to be susceptible to organophosphorus larvicides. Those oils which displayed high biological activities against the laboratory strain were evaluated further against fourth instar larvae of *Aedes nigromaculis* collected from the Galanis pasture in Merced County.

Methods utilized for the evaluation of petroleum oils against mosquito larvae in the laboratory were described by Darwazeh et al. (1972). Twenty-five fourth instar larvae were placed in 800 cm<sup>2</sup> white enamel pans containing 2,000 ml of tap water. Petroleum oils were applied with a microsyringe. Mortality readings were taken at 4, 8, and 24 hours after treatment. All pans were lined with aluminum foil to avoid contamination and were rinsed with hot water followed by acetone after each test.

Petroleum larvicidal oils were tested against the mosquito fish *Gambusia affinis*. The method of bioassay utilized in these studies was as described above for the bioassay of mosquito larvae. In Test 1, 10 fish (0.8-1.5 inches in length) were placed in a pan containing 2,000 ml of tap water. In Test 2 the amount of water was increased to 4,000 ml per pan to reduce the concentration of oil from 160 to 80 ppm.

Field evaluations were conducted in the Monteiro pasture, Tulare County; the Smith pasture, Kern County; and the Edna pasture, Merced County. All test applications were made by aircraft belonging to the respective mosquito abatement districts.

The Tulare tests were applied with a Piper Pawnee PA-25 equipped with ten D-6 and nine D-8 nozzles, delivering one gal/acre at 90 mph over an assumed swath width of 60 feet. In order to apply 2 and 3 gal/acre each swath was flown two and three times. The Kern tests were applied with a Callair A-9B equipped with 14 D-6 nozzles, delivering one gal/acre at 90 mph over an assumed swath width of 60 feet with a pressure of 40 psi. To apply two gal/acre the number of nozzles was increased to 27 while other conditions remained unchanged. The Merced tests were applied with a Callair A-9B equipped with 26 size 8010 nozzles, delivering two gal/acre at an assumed swath width of 40 feet.

In the first Tulare tests, Chevron Research 72R-2569 and Chevron Research 72R-2570 were applied at two gal/acre while Chevron Research 72R-2593 was applied at 2 and 3 gal/acre. In the second tests, Chevron Research 72R-2569 and Golden Bear 1313 were applied at two gal/acre. Each test plot consisted of 15 acres of irrigated pasture with a dense 6 to 8 inches of vegetation cover. The water was widespread and 4-6 inches deep. Test larvae were third and fourth instar *A. nigromaculis* known to be organophosphorus resistant (Table 1). Larval counts (25 dips/plot) were taken prior to treatment and 24 hours post-treatment in the first test, while post-treatment counts were made at 4, 24, and 48 hours in the second test.

In the Kern County tests Chevron Research 72R-2569 was evaluated at 1 and 2 gal/acre. The field vegetation was moderately dense and 4-6 inches high and the water depth was 4-6 inches. Both fourth instar larvae and pupae of *A. nigromaculis* were present. This population is moderately resistant to parathion and methyl parathion and susceptible to other organophosphorus larvicides (Table 1). Larval counts were taken prior to treatment and 4 and 24 hours post-treatment.

In the Merced County tests Chevron Research 72R-2569 and Golden Bear 1313 were applied at two gal/acre. Each material was applied to portions of the field with open water and dense vegetation. Third and fourth instar *A. nigromaculis* larvae are known to be organophosphorus resistant (Table 1). Counts were made in the open water and heavily vegetated areas before treatment and 4, 14, and 48 hours post-treatment.

**RESULTS AND DISCUSSION.**—Table 2 lists biological activity of several petroleum oils against *C. p. quinquefasciatus* in the laboratory. All materials showed high activity at rates of one gal/acre and less at 0.5 and 0.25 gal/acre. Laboratory measurements showed that the compounds at one gal/acre were somewhat less active against fourth instar *A. nigromaculis* (Table 3).

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Table 1.—Susceptibility levels of fourth instar *Aedes nigromaculis* larvae to various organophosphorus larvicides in plots used for the evaluation of petroleum oils, July-August, 1972.

Source	Pasture Identification	Malathion		Methyl Parathion		Parathion		Dursban		Fenthion	
		LC <sub>50</sub>	LC <sub>90</sub>	LC <sub>50</sub>	LC <sub>90</sub>	LC <sub>50</sub>	LC <sub>90</sub>	LC <sub>50</sub>	LC <sub>90</sub>	LC <sub>50</sub>	LC <sub>90</sub>
Kern	Smith	0.045	0.075	0.042	0.18	0.095	0.15	0.002	0.005	0.0035	0.009
Tulare	Montiero	0.110	0.310	0.29	0.63	0.37	0.98	0.005	0.02	0.025	0.046
Merced	Edna	0.02	0.05	0.036	0.37	0.140	0.75	0.0098	0.03	0.016	0.08

Table 2.—Laboratory evaluation of petroleum oils against *Culex pipiens quinquefasciatus* larvae, February-March, 1972.

Petroleum Oil	Dosage		Average (percent) Mortality of Indicated Larval Stages		
	gal/A	ul/cm <sup>2</sup>	1-2	3-4	P
Chev. Res. 72R-2569	0.25	0.025	35	53	--
	0.50	0.050	83	95	100
	1.00	0.100	100	100	100
Chev. Res. 72R-2571	0.25	0.025	--	18	--
	0.50	0.050	--	80	--
	1.00	0.100	--	100	--
Kern MAD-492	0.25	0.025	--	55	--
	0.50	0.050	--	90	--
	1.00	0.100	--	100	--
Golden Bear 1313	0.25	0.025	--	28	--
	0.50	0.050	--	48	--
	1.00	0.100	--	93	--
Check	--	--	0	0	0

Table 3.—Laboratory evaluation of petroleum oils against fourth instar *Aedes nigromaculis* larvae at the rate of 1.0 gal/a., August, 1972.

Petroleum Oil	Avg. (%) mortality after treat.(hrs)		
	4	8	24
Chev. Res. 72R-2569	70	74	94
Golden Bear 1313	4	4	60
Kern MAD-492	66	82	82
Flit MLO	8	16	40
Check	0	0	0

Tables 3, 5, and 6 present the results of field evaluations in Tulare, Kern, and Merced counties. More information was obtained with Chevron Research 72R-2569 than with any of the other materials. At the rate of two gal/acre, Chevron Research 72R-2569 achieved 100% control in all areas at 24 hours post-treatment. It appeared, however, that earlier kills occurred in the areas where organophosphorus resistance was less severe. Golden Bear 1313 did not show the rapid initial knockdown of Chevron Research 72R-2569, but kills were comparable (though lower) at 24 and 48

hours post-treatment. Table 7 shows the results of tests against mosquito fish. Golden Bear 1313 and Flit MLO® were apparently harmless to *G. affinis* when applied at 2 and 4 gal/acre (80 and 160 ppm, respectively) in the laboratory. Chevron Research 72R-2569 at two gal/acre (80 ppm) produced no mortality, but in one test at four gal/acre (160 ppm) 40% mortality occurred.

#### Acknowledgment

"The assistance of Michael Gutierrez, California Department of Public Health, Bureau of Vector Control and Solid Waste Management, and the cooperation of Kings, Kern, Merced and Tulare Mosquito Abatement Districts' managers and employees is acknowledged."

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Table 4.--Field evaluation of petroleum oils for the control of fourth instar *Aedes nigromaculis* larvae in irrigated pastures, Tulare County.

Petroleum Oil	Dosage gal/a	Average larvae / dip							
		Pre-treat.	Post-treatment (hours)				%Reduction after Treatment (hrs)		
			4	24	48	4	24	48	
<b>(Test I - 7/7/72)</b>									
Chev. Res. 72R-2570	2.0	20							
Chev. Res. 72R-2570	2.0	20	--	1	--	--	95	--	
Chev. Res. 72R-2593	2.0	35	--	0.4	--	--	99	--	
Chev. Res. 72R-2593	3.0	27	--	0	--	--	100	--	
Chev. Res. 72R-2569	2.0	21	--	0	--	--	100	--	
Check	--	12	--	23	--	--	0	--	
<b>(Test II - 8/21/72)</b>									
Chev. Res. 72R-2569	2.0	33	10	0.2	0	70	99	100	
Golden Bear 1313	2.0	27	29	3.5	1.5	0	87	94	
Check	--	21	26	25	28	0	0	0	

Table 5.--Field evaluation of petroleum oil (Chevron Research 72R-2569) for the control of fourth instar larvae and pupae of irrigated pasture mosquito, *Aedes nigromaculis*, Kern County, July 26, 1972.

Dosage (gal/a)	Average number of larvae and pupae/dip							
	Pre-treatment		Post-treatment (hours)				Total % reduction after treatment (hours)	
	L	P	4		24		4	24
2.0	53	1	6*	0*	0.2*	0.1*	89	99.5
1.0	58	4	37	0	10.0	8.0	40	70.0
Check	21	1	23	1	27.0	3.0	0	0
2.0	27	8	0.35*	0*	0.0*	0.0*	99	100
Check	29	5	27	8	17.0	19.0	0	0

\*Dead larvae and pupae recovered by dipping.

Table 6.--Field evaluation of petroleum oils for the control of fourth instar *Aedes nigromaculis* larvae in irrigated pastures, Merced County, August 22, 1972.

Petroleum Oils	Dosage gal/a	Average number of larvae/dip						
		Pre-treat.	Post-treatment (hours)			% reduction after treatment (hrs)		
			4	24	48	4	24	48
<b>(Open Water)</b>								
Chevron 72R-2569	2.0	20	4	0	0	80	100	100
Golden Bear 1313	2.0	52	58	8	1	1	85	98
<b>(Heavy Vegetation)</b>								
Chevron 72R-2569	2.0	7	1	0.35	0	86	95	100
Golden Bear 1313	2.0	37	38	2	0.5	0	90	99
Check	--	15	16	18	17	0	0	0

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Table 7.—Effect of petroleum oils against the mosquito fish, *Gambusia affinis*, in the laboratory, August 24, 1972.

Petroleum Oil	Dosage		Avg. % mortality
	gal/a	ppm	
(Test I <sup>a</sup> )			
Chevron 72R-2569	2.0	80	0
Chevron 72R-2569	4.0	160	40
Flit MLO	2.0	80	0
Flit MLO	4.0	160	0
Golden Bear 1313	2.0	80	0
Golden Bear 1313	4.0	160	0
Check	--	--	0
(Test II <sup>b</sup> )			
Chevron 72R-2569	2.0	40	0
Chevron 72R-2569	4.0	80	0
Check	--	--	0

<sup>a</sup>Pans contained 2,000 ml water

<sup>b</sup>Pans contained 4,000 ml water

# SUSCEPTIBILITY LEVEL OF ORGANOPHOSPHORUS RESISTANT *Aedes nigromaculis* MOSQUITO LARVAE TO PETROLEUM OILS

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**INTRODUCTION.**—Previous field evaluations of petroleum oils against organophosphorus multiple-resistant mosquito larvae in irrigated pastures were discouraging. Gillies et al. (1971) and Schaefer and Ramke (1971) were unable to achieve consistent larval control with Flit MLO® when applied by air at the rate of 2.0 gallons per acre. Darwazeh et al. (1972) and Darwazeh and Ramke (1972) achieved excellent control with petroleum oils in Kings County but the same oils at the same rates failed to render measurable control in Tulare County.

Schaefer and Ramke (1971) attributed the variability in results obtained to problems of uneven dispersal of the oil across the aircraft swaths. Preliminary laboratory investigations by Darwazeh et al. (1972) indicated Tulare pasture mosquito larvae to be more tolerant than those in Kings County to petroleum oils. In order to test these findings further, the studies documented here were undertaken to determine the toxicity level of petroleum oils against mosquito larvae with a known susceptibility level to various organophosphorus mosquito larvicides.

**METHODS AND MATERIALS.**—*Aedes nigromaculis* larvae were collected from irrigated pastures in Madera, Kern, Tulare, Merced, Kings, and Fresno counties. The larvae then were tested in the laboratory against MLO® and a newly developed material (Chevron Research 72R-2569) at the rate of 0.5 and 1.0 gallons per acre. Flit MLO is currently being used successfully by many mosquito control agencies in California. Chevron 72R-2569 achieved excellent control against mosquito larvae of various susceptibility levels to organophosphorus larvicides. Applications were made by aircraft at the rate of 2.0 gallons per acre (Darwazeh 1973). Comparative tests were also run to determine the susceptibility level of the larvae to parathion, methyl parathion, fenthion, malathion, and chlorpyrifos.

The petroleum oil tests were conducted in 800 cm<sup>2</sup> white enamel pans. The pans were lined with aluminum foil and 20 fourth instar larvae were placed in each pan containing 2,000 ml of water. The petroleum oils were applied with a Calab microsyringe, 2500 micrometer unit, and mortality readings were obtained 24 hours after treatment. To determine the susceptibility level of mosquito larvae to organophosphorus larvicide a procedure was employed as described by Mulla et al. (1966). A 1% stock solution (weight/volume) of the technical grade of each compound was prepared in acetone and several dilutions were made as

needed. Aliquots of 1 ml or less of the proper strength solution were added to 4 oz waxed paper cups containing 20 fourth instar larvae in 100 ml of tap water. After 24 hours of exposure, mortality readings were taken and LC<sub>50</sub> and LC<sub>90</sub> estimates were obtained graphically.

**RESULTS AND DISCUSSION.**—As previous studies indicated, mosquito larvae susceptible to organophosphorus larvicides were found to be susceptible to low application rates of the petroleum oils tested. Against the susceptible strain, complete mortality occurred with Chevron Research 72R-2569 while 53%-64% mortality was obtained with Flit MLO in the laboratory at the rate of 1.0 gallon per acre (Tables 1-3).

Complete mortality was impossible to achieve against the highly resistant larvae with any oil tested at the rate of 1.0 gallon per acre. Mortality varied according to the susceptibility level of the larvae to the organophosphorus larvicide as well as to the number of organophosphorus larvicides the mosquito can tolerate. Larvae collected from the Monterey pasture were highly resistant to all chemicals, with the LC<sub>90</sub> to methyl parathion being 2.0 ppm. Against these larvae, no mortality occurred with Flit MLO and only 24% was obtained with Chevron Research 72R-2569 at the rate of 1.0 gallon per acre. At the same rate, no mortality occurred with Flit MLO but 45% was achieved with Chevron 72R-2569 against the McHenry mosquito larvae where the LC<sub>90</sub> to methyl parathion was 1.7 ppm.

In conclusion, mosquito larvae susceptibility level to organophosphorus larvicides is a factor to be considered in determining the application rate of petroleum oils by aircraft for satisfactory and consistent results.

## Acknowledgment

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Table 1.—Twenty-four-hour susceptibility level of fourth instar *Aedes nigromaculis* larvae to organophosphorus larvicides and petroleum oils from various areas, 1972. S = susceptible, M = moderately resistant, R = resistant.

Mosquito Abatement District	Pasture Identification	Petroleum Oils			OP Larvicides --- LC <sub>90</sub> (ppm)				
		Dosage (gal/a)	% Mortality		Malathion	Parathion	MParathion	Fenthion	Dursban
			Flit MLO	Chev. 72R-2569					
Uncontrolled Tulare Co.	Gilbert, Porterville	0.5	33	75	0.1	0.02	0.007	0.005	0.0027
		1.0	53	100	S	S	S	S	S
Madera	Pistoresi	0.5	--	52	--	0.007	0.007	0.01	0.001
		1.0	--	100	--	S	S	S	S
Madera	Huntley	0.5	18	40	--	0.06	0.025	0.0035	0.0015
		1.0	35	95	--	M	S	S	S
Kern	Smith	0.5	40	54	--	0.11	0.097	0.007	0.0048
		1.0	64	86	--	R	M	S	S
Merced	Edna	0.5	18	18	0.05	0.75	0.37	0.084	0.032
		1.0	18	70	S	R	R	R	R
Merced	Lewis Bros.	0.5	10	52	0.17	0.25	0.25	0.06	0.018
		1.0	22	68	S	R	R	R	R
Fresno West-side	Muller	0.5	0	34	0.26	1.5	0.07	0.17	0.06
		1.0	0	68	M	R	R	R	R
Fresno West-side	McHenery	0.5	0	0	0.58	1.40	1.70	0.5	0.104
		1.0	0	45	R	R	R	R	R

Table 2.—Susceptibility level of fourth instar *Aedes nigromaculis* larvae to organophosphorus larvicides and petroleum oils, Kings, County, 1972. S = susceptible, M = moderately resistant, R = resistant.

Pasture Identification	Petroleum Oils			OP Larvicides --- LC <sub>90</sub> (ppm)				
	Dosage (gal/a)	% Mortality		Malathion	Parathion	MParathion	Fenthion	Dursban
		Flit MLO	Chev. 72R-2569					
Nunes	0.5	--	68	>0.1	>0.03	>0.03	>0.003	0.0014
Nunes	1.0	--	100	S	S	S	S	S
Espinela Bros.	0.5	12	50	0.2	0.069	0.064	0.0073	0.0024
Espinela Bros.	1.0	36	95	M	M	M	S	S
Haker, S.	0.5	--	42	0.12	0.07	0.14	0.007	0.004
Haker, S.	1.0	--	100	S	M	R	S	S
Schieringa, S.	0.5	--	54	0.3	0.13	0.08	0.014	0.003
Schieringa, S.	1.0	--	96	M	R	M	S	S
Costa	0.5	--	24	0.28	0.14	0.29	0.007	0.003
Costa	1.0	--	96	M	R	R	S	S
Cordoza, F.	0.5	0	50	0.23	0.175	0.40	0.012	0.007
Cordoza, F.	1.0	0	90	M	R	R	S	M
Katen, A.	0.5	--	42	0.4	0.23	0.14	0.01	0.004
Katen, A.	1.0	--	77	R	R	R	S	S



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Table 3.—Susceptibility level of fourth instar *Aedes nigromaculis* larvae to organophosphorus larvicides and petroleum oils, Tulare County, 1972. S = susceptible, M = moderately resistant, R = resistant.

Pasture	Petroleum Oils			OP Larvicides (LC <sub>90</sub> ) ppm				
	Dosage	% Mortality		Malathion	Parathion	MParation	Fenthion	Dursban
Identification	(gal/a)	Flit MLO	Chev. 72R-2569					
Montiero, M.	0.5	0	0	0.3	1.14	2.0	0.09	--
	1.0	12	26	R	R	R	R	--
Cordoza, L.	0.5	0	24	0.2	0.5	0.39	0.02	0.005
	1.0	2	78	M	R	R	M	S
Smith, J.	0.5	6	26	0.45	0.38	0.82	0.0159	0.005
	1.0	12	86	R	R	R	S	S
Hoffman	0.5	--	66	0.16	0.18	0.25	0.013	0.004
	1.0	--	95	S	R	R	S	S

# MEASUREMENT OF PROPOXUR RESISTANCE IN ADULT *Aedes nigromaculis* BY TIME EXPOSURE — A PROGRESS REPORT

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**INTRODUCTION.**—The pasture mosquito, *Aedes nigromaculis*, has been under larvicide pressure for many years and larvae have become highly resistant to many organophosphorus chemicals (Womeldorf et al. 1971). Consequently, an increasing number of mosquito control agencies are reducing their larviciding operations and depending more upon adulticides. Organophosphorus compounds have failed to control adult *A. nigromaculis* in resistant areas, but the carbamate propoxur was used to re-establish control (Ramke et al. 1969). However, in 1970 suspected control failures with propoxur were reported in a pasture in the Turlock Mosquito Abatement District.

A program to monitor developing propoxur resistance in California adult mosquitoes would confirm or deny high adult tolerance in the event of an observed operational failure or warn against approaching problems due to incipient resistance. Many workers have provided estimates of median lethal dosage of propoxur for various California adult strains. The technique calls for exposure of the insects to a range of concentrations for a constant time (Georghiou and Metcalf 1961). With this technique, Schaefer and Wilder (1970) found only a 1.6-fold difference in the median lethal dosage between susceptible and operationally-resistant adult *A. nigromaculis*, which we considered inadequate to assure clear differentiation. At the suggestion of Dr. R. Pal (personal communication) it was postulated that exposing susceptible and resistant strains to equal dosages of propoxur, but for a range of time periods, might elucidate the difference between susceptible strains and those that were difficult to control in the field. This report presents preliminary results of laboratory tests to measure propoxur resistance in adult *A. nigromaculis* by a variable time-exposure technique.

**MATERIAL AND METHODS.**—**Rearing Method.**—*A. nigromaculis* is not easily colonized, consequently all tests were conducted with adults reared from field-collected larvae. About 200 to 300 larvae or pupae were collected in plastic-lined, ½-gallon ice cream cartons. Water from the collection site was used. A one-gallon ice cream carton with a screened top, a cut-out bottom and a stoppered hole on the side was subsequently snapped on the ½-gallon carton. This assembly was held in a room at 23.4 - 25.7°C (72° - 74°F). Wet raisins were placed on top of the screen for nutriment. After emergence the adults moved to the large carton where they could be aspirated through the hole on the side. The water in the smaller carton kept the humidity at an adequate level inside the cage and no abnormal mortality was observed. Age control was facilitated by the relatively small numbers handled per cage.

**Strains Utilized.**—The susceptible strain for these tests was collected from a pasture situated outside the insecticide-controlled area of the Tehama County Mosquito Abatement District and is herein referred to as the Bend strain. The resistant strain was collected from a pasture in the Turlock Mosquito Abatement District, and has resisted control by aerial applications with .05 lb/acre of propoxur. It is known as the Hatch strain. Multiple resistance to organophosphorus larvicides has been shown in field treatments as well as in laboratory tests.

**Insecticide Testing Technique.**—An objective of all tests was to establish lethal dosage (LD) and lethal time (LT) regression lines by utilizing three or more dosages or time exposures of the chemical propoxur. Both dosage and time-exposure treatments were run simultaneously with the same generation where possible. All tests were run in duplicate.

The testing procedure was essentially as described by Georghiou and Metcalf (1961) with the modification of Darwazeh (1972). Fifteen to 20 adult mosquitoes were aspirated from the rearing cage into the vial. The sexes were not separated and the females were not blood-fed. Insecticide treated filter paper (Whatman No. 2) was then placed into the test vial which was then stoppered. After the appropriate time-exposure periods, the mosquitoes were transferred into an eight-ounce paper cup and held for 24 hours. Mortality was determined thereafter. The holding cage contained a cotton wick saturated with 5% sugar water providing food and moisture.

**Data Analysis.**—Probit regression lines were fitted to the data using the computer program of the Data Processing Center of the State Department of Public Health. The program includes a statistical test of the homogeneity or regularity of each set of data in terms of how well it fits a probit line. Only tests for which an acceptable fit was obtained were included in the analysis.

**RESULTS AND DISCUSSION.**—From values of the probit lines (Table 1) and examination of the raw data, the following was observed:

1. Of the three tests on "susceptibles" (S), the June test shows a steeper slope and lower  $LT_{50}$  than the September tests,
2. There is some evidence of a plateau in the upper LT range for the "resistant" (R) mosquitoes,
3. The LD and LT confidence limits multiplied by the constant time and concentration, respectively, are reasonably consistent and indicate approximately 2½ fold greater tolerance in the R population.
4. There is overlap for S and R in both time and dosage ranges

These observations were made from rather limited data, and although some of the peculiarities of the slopes are discussed, no tests of the statistical significance of differences of the slopes have been run.

It was noted that an S test, run in June, yielded a steeper slope and a lower  $LT_{50}$  than the September test. The difference between the June test and the highest September test is 7.8 minutes. If the differences continue to be substantial in future tests, it may be necessary to establish separate baselines for each month that the populations are tested.

There was some indication of a plateau in the upper LT range. R. Georghiou et al. (1966) working with *Culex fatigans* selected with propoxur, also observed such a plateau. They proved that behavioristic avoidance of the insecticide was not the cause, but suggested as a likely explanation an enhanced rate of metabolism of the insecticide, the effect of which would be magnified by reduction of tarsal absorption of the toxicant. The same may be true for our tests with *A. nigromaculis*.

Assuming that the effective dose equals concentration multiplied by exposure time, the confidence limits from the LT and LD tests may be compared (see Table 1). For susceptible strain the  $LT_{50}$  yields  $4.2(17.4 - 31.8) = 73.08 - 133.56$  compared to  $60(1.26 - 1.67) = 75.60 - 100.20$  from the  $LD_{50}$ . For the resistant strain the  $LT_{50}$  yields  $4.2(41.4 - 64.8) = 173.88 - 272.16$  compared to  $60(3.61 - 4.72) = 216.60 - 283.20$  from the  $LD_{50}$ . These results are reasonably consistent and indicate approximately 2½ fold greater tolerance in the resistant population.

A 5% to 95% mortality range for these strains can be expected if future tests are carried out using  $4.2 \mu\text{g}/\text{cm}^2$  of propoxur, as a standard concentration, and equal log exposure time intervals for S between 10 and 160 minutes for R between 20 and 320 minutes. Effective LD ranges can be achieved by using dosages between .83 and  $6.90 \mu\text{g}/\text{cm}^2$  for S; a dosage range between 2.80 and  $8.30 \mu\text{g}/\text{cm}^2$  for R. Overlap of the LT and LD ranges for S and R is unavoidable.

At this point it is not possible to conclude if the LT method renders a clearer differentiation between susceptible and operationally resistant adults than the LD method. Further tests with the above defined ranges and comparison of the results are necessary. As more data become available, it should also be possible to determine a failure threshold for propoxur against *A. nigromaculis* adults.

**SUMMARY.**—*Aedes nigromaculis* from a resistant area and from an uncontrolled area were tested with the variable exposure time technique. By using  $4.2 \mu\text{g}/\text{cm}^2$  propoxur as a standard concentration and exposing the susceptible adults at 10, 20, 30, 80, 150 minutes and the resistant adults at 20, 40, 80, 160, 320 minutes, a 5% to 95% mortality range can be expected. More data will be compiled in the future to determine the failure threshold.

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Table 1.—Probit line values of bioassays of propoxur against adult *Aedes nigromaculis* — California, 1972. (S = susceptible, Bend strain; R = resistant, Hatch strain.)

A. Constant concentration $4.2 \mu\text{g}/\text{cm}^2$ - variable exposure time in minutes.									
Date	Strain	Slope	$LT_{50}$	95% Confidence limits		$LT_{90}$	95% Confidence limits		$CT_{50}$ (Conc x time)
6-21-72	S	7.02	21.0	18.6	- 23.4	31.8	28.2	- 38.4	88.2
9-13-72	S	3.02	28.8	24.6	- 33.0	76.8	62.4	- 105.0	120.96
9-20-72	S	3.81	24.0	17.4	- 31.8	51.6	37.8	- 97.80	100.8
9-20-72	R	2.82	53.4	41.4	- 64.8	151.2	117.6	- 229.2	224.28

B. Variable concentrations ( $\mu\text{g}/\text{cm}^2$ ) - constant exposure time 60 minutes.									
Date	Strain	Slope	$LT_{50}$	Limits		$LD_{90}$	Limits		$CT_{50}$ (Conc x time)
9-13-72	S	3.58	1.4	1.26	- 1.67	3.33	2.80	- 4.20	84.
9-20-72	R	5.39	4.2	3.61	- 4.72	7.36	6.39	- 9.17	252.

# EFFECTIVENESS OF A FLIT MLO® – LARVICIDE OIL FORMULATION IN THE FIELD

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Southeast Mosquito Abatement District  
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The Southeast Mosquito Abatement District has been using Flit MLO® since 1968. However, it was not until 1971 that operational use of Flit MLO was started. Prior to that time, Flit MLO was used in water impoundments such as swimming pools, water troughs, ornamental ponds, and other spot-treatment sites. In most of these situations, a high level of control was obtained.

In 1971, when operational use of Flit MLO was started with spraying of street gutters, catch basins, and flood control channels, it was observed that a marked reduction in the level of control occurred in these types of mosquito sources. Since Flit MLO has shown good pupicidal properties in laboratory and field tests and also displays low phytotoxicity and is not corrosive at recommended dosages, it was felt that Flit MLO should not be abandoned until it was given a fair trial. Also, three of the pest species of mosquitoes have started to show a high level of resistance to organophosphorous compounds in many areas of the District. Consequently, the use of alternate larvicides is becoming more important.

It was observed early in the work with Flit MLO, that vegetation or other material in the water seemed to effect the level of control in a given breeding site by breaking the continuous film of oil on the surface. Open spaces around vegetation would occur, allowing some of the mosquito larvae to seek these out and survive. Since oils are surface active materials, it is obvious that movement of water would tend to diminish the level of control in a given mosquito source. The movement of water in flood control channels, rivers, and streams precludes the use of surface-active materials presently known today. This was the experience obtained with Flit MLO in street gutters. During the 1971 season, the control operators noticed mosquito larvae and pupae still alive in gutter water sprayed one to two days prior to post-treatment inspection. Many service requests were received from residents in areas that had been treated with Flit MLO on a routine basis every seven to eight days.

However, in street gutters the water movement is periodical during the day so correct timing of the treatment could yield results if the "knockdown" or killing time is sufficiently short. In order to decrease the killing time, another oil was added to the Flit in sufficient quantities to shorten the killing time without unduly increasing the phytotoxicity or corrosive qualities. By "killing time" it is meant the time it takes from application to the time the organism has received sufficient oil to completely inactivate it so as not to allow recovery.

The purpose of this study was to evaluate the effectiveness of a Flit MLO larvicide oil combination compared to straight Flit MLO under field conditions.

**MATERIALS AND METHODS.**—A study site was chosen in the Long Beach area which had a long history of mosquitoes breeding in the street gutters. The test area was approximately ½ mile square, with 14 miles of gutters. The parkways are heavily vegetated with large trees, providing leaf litter to obstruct the flow of water in many areas.

Larval sampling consisted of three sample sites to be monitored on a weekly basis in addition to pre- and post-treatment samples. The samples consisted of three dips composited per sample site. The samples were returned to the laboratory where the larvae were identified and counted. All stages were counted pre- and post-treatment.

The amount of actual water sprayed in the 14 miles of gutters averaged 6 miles. The total area sprayed was approximately 0.8 acres. Dosage rate averaged 5 gallons of oil mixture per acre.

The following oils were used in the study project: 1) Flit MLO plus Richfield Larvicide "A" at a 4-1 ratio; 2) Flit MLO without additive. Both oils were used at the same dosage rate of 5 gallons per acre.

The test area was normally sprayed on Monday mornings at approximately 11:00 a.m. The movement of water in the gutters was variable, but the mornings averaged less movement than the afternoons.

Equipment used for treatment of the test area consisted of a Jeep postal van (right-hand drive) equipped with a 55-gallon tank, an electric pump with a spray tip that delivered material at the rate of 0.1 gpm (gallon per minute) at a driving speed of 5-7 miles per hour.

Table 1.—Represents the average number of larvae and pupae indicating the total percent reduction received with the Flit MLO – Richfield Larvicide "A" combination compared to Flit MLO unfortified.

Average number of larvae (all stages) and pupae per dip			
Material	Pre-Treatment	Treatment	Percent Reduction
Flit MLO + Richfield Larvicide "A" <sup>1</sup>	21.6	2.4	88.0
Flit MLO <sup>2</sup>	13.5	11.1	17.7

<sup>1</sup> Average of 7 replicates

<sup>2</sup> Represents one treatment

Experiments involving the Flit MLO - Richfield Larvicide "A" combination were replicated seven times. One experiment was performed using Flit MLO without fortification.

The mosquito species most prevalent in the street gutters during the test period was *Culex pipiens quinquefasciatus*, but *Culex peus* and *Culex tarsalis* were occasionally found.

**RESULTS AND DISCUSSION.**—The fortified Flit MLO mixture was clearly superior to the Flit MLO (unfortified) formulation, as shown in Table 1. However, the average reduction that occurred was below the level of control we strive to obtain in our control program. The data on Flit MLO (unfortified) was inconclusive and can only be used for comparative purposes as only one experiment was performed.

Table 2 presents a breakdown on the percent reduction obtained regarding the immature stage of development. From this data, one can see that fortified Flit MLO gave good reduction of 4th instar larvae with Flit MLO (unfortified) producing poor results. Flit MLO - Richfield Larvicide "A" combination also better killed the immature larvae (1st and 3rd instar) than the Flit MLO (unfortified). The phenomenon of high paraffinic oils not killing early instar larvae at the same levels as required for 4th instar larvae and pupae seems to exist even with the fortified Flit MLO, but the level of reduction was higher compared to Flit MLO (unfortified) as shown in Table 2.

Figure 1 shows average larval and pupal population levels on a weekly basis for the period of the study. The high peak observed for the last of July resulted from not treating the study area for two weeks to allow the larval population to build up again for one of the experiments. From this graph, one will notice that even though there was weekly treatment except for one or two instances, an extremely high level of control was not achieved. Adult mosquitoes were being produced indicating that complete adult emergence suppression was not occurring. However, the adult emergence was reduced considerably as evidenced by a 38% reduction in service requests in the study area over the number received in 1971 for the same period of time.

In summary, it can be said that the fortified Flit MLO gave better control during the peak portion of the season in our street gutters than did Flit MLO (unfortified). However, this material still did not provide the level of control we would like to achieve.

This study will be continued in 1973 using the same oils in addition to some new oils with different additives.

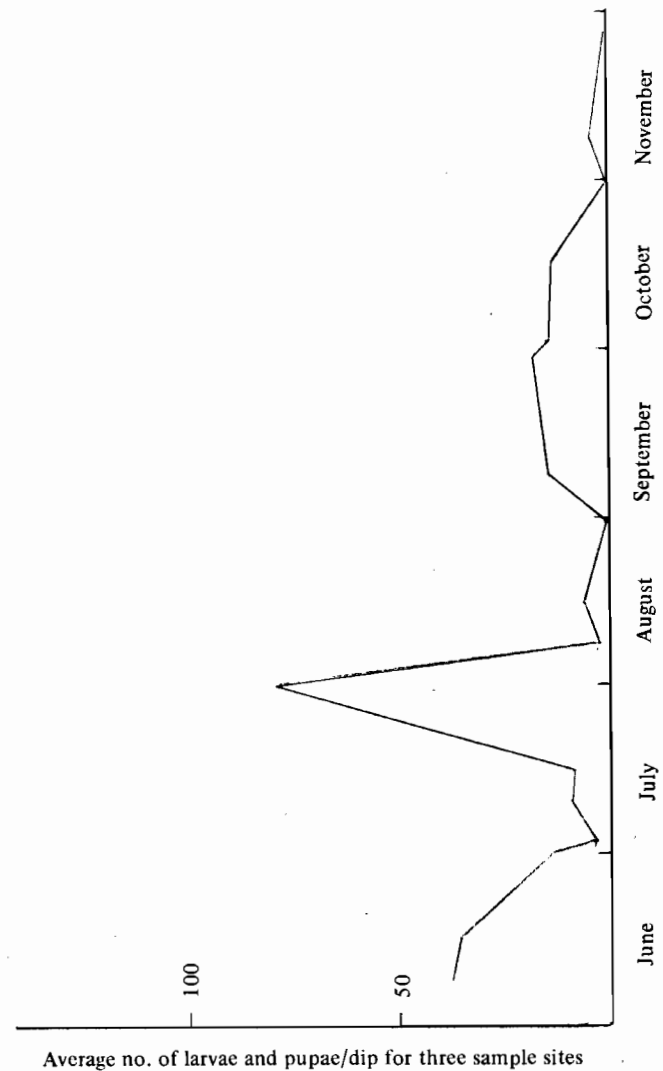


FIG. 1.—Average pre-treatment population of larvae and pupae of the three sample sites recorded over the period of the test.

Table 2.—Stage of immature mosquito development and the percent reduction received for Flit MLO - Richfield Larvicide "A" combination compared to Flit MLO unfortified.

Material	Average number 4th instar, immature larvae and pupae per dip				% Reduct.	Immature	% Reduct.	Pupae	% Reduct.
	4th instar	Immature <sup>1</sup>	Pupae	4th instar					
Flit MLO + Richfield Larvicide "A"	5.7	11.4	4.5	0.43					
Flit MLO + Richfield Larvicide "A"	5.7	11.4	4.5	0.43	02.94				
Flit MLO + Richfield Larvicide "A"	5.7	11.4	4.5	0.43	92.4	1.9	83.4	0.11	97.6
Flit MLO	1.2	11.7	0.66	1.7	0	7.7	34.1	1.70	0

<sup>1</sup>immature larvae = 1st 3rd instar