

potential means of control. Field trials of such applications are necessary to determine the acceptance of JH baits presented in competition with the food normally available.

**ACKNOWLEDGMENTS.** The author thanks Mrs. Virginia J. Brown for technical assistance in these studies. Recognition is also due Mr. William E. Dale and Miss Mary Goette for preparing the treated graham flour. These studies were accomplished as part of a contractual agreement between the Center for Disease Control and the Agency for International Development.

#### Literature Cited

- Jakob, W. L. and Schoof, H. F. 1971. Studies with juvenile hormone-type compounds against mosquito larvae. *Mosq. News* 31(4):540-543.
- Jakob, W. L. and Schoof, H. F. 1972. Mosquito larvicide studies with MON 585, a juvenile hormone mimic. *Mosq. News* 32(1):6-10.
- Sacher, R. M. 1971. A mosquito larvicide with favorable environmental properties. *Mosq. News* 31(4):513-516.
- Schaefer, C. H. and Wilder, W. H. 1972. A practical evaluation of insect developmental inhibitors as mosquito control agents. *J. Econ. Entomol.* 65(4):1066-1071.
- Staal, G. B. 1971. Practical aspects of insect control by juvenile hormone. *Bull. WHO* 44:391-394.
- Wilton, D. P., Fetzer, L. E., Jr. and Fay, R. W. 1972. Quantitative determination of feeding rates of *Anopheles albimanus* larvae. *Mosq. News* 32(1):23-27.

## SOME INFLUENCES OF AQUATIC VEGETATION ON THE SPECIES AND NUMBER OF CULICIDAE (DIPTERA) IN SMALL POOLS OF WATER

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**ABSTRACT.** Physical configuration of the aquatic vegetation in 10 ft. diameter plastic pools in the southeastern United States appeared to influence qualitative parameters of mosquito populations. There were definite trends reflecting the association of particular species of Culicidae with a particular type of aquatic plant: submerged, floating, emergent or none. Even though apparent quanti-

tative differences were observed in culicid populations among the various types of vegetation, vegetation had no significant effect on population sizes. The floating plant, *Spirodela oligorrhiza* (duck weed), had a detrimental effect on mosquito survival when it completely covered the surface of the water.

**INTRODUCTION.** Most mosquito breeding is associated with aquatic vegetation (Bates, 1949). Thus, understanding the mosquito-aquatic plant relationship is important. Plants may influence mosquitoes in many ways. They may physically restrict oviposition either by disrupting the behavior of the female or by rendering the surface of the water inaccessible (Macan and Worthington, 1951). Conversely, metabolites or biochemical processes of plants may act as attractants to the female, leading her to favorable oviposition habi-

tats (Rapp and Emil, 1965). Happold (1965) observed that the distribution of mosquito larvae in nature was influenced by the female's behavioral reaction to vegetation, amount of shade, and habitat location.

The influence of plants is important to both adult and immature mosquitoes. Aquatic vegetation influences many of the physiochemical and ecological relationships that contribute to the success of immature forms. Plants reduce wave action of the water and thus provide a more

stable interface for the exchange of gases (Rueger *et al.*, 1964); conceal and protect the eggs, larvae, and pupae from predators (Bradley, 1932); provide shade from intense solar radiation (Zetek, 1920); and provide a substrate for algae and other larval food (Barber and Hayne, 1925). Plants also influence water temperature, evaporation rate, chemical content, oxygen supply and attachment sites for food organisms (Horsfall, 1967). Certain products of plant metabolism, especially plant growth hormones, promote survival of certain mosquito species (Abdel-Malek, 1948).

However, some plants inhibit the survival of immature mosquitoes. Among these, the most unusual are carnivorous plants, such as *Utricularia macrorhiza*, that capture and consume mosquito larvae (Twinn, 1931). Caballero (1919) reported larvicidal or antimosquito properties of several species of Characeae, but MacGregor (1924) found the same species incapable of deterring mosquito breeding. Some species of blue-green algae limit mosquito production in rice fields (Gerhardt, 1954). Complete surface mats of floating plants also limit mosquito breeding (Smith, 1910). Bradley (1932) attributed the effectiveness of complete mats of *Lemna* in decreasing *Anopheles* spp. breeding to three factors: (1) larval food supply is decreased because phytoplankton do not flourish, (2) larvae cannot break the surface film for respiration, and (3) larvae are not effectively hidden by the plants from enemies in the water. Smith and Enns (1967) also noted that mosquito breeding was virtually eliminated in an oxidation pond in Missouri which was covered by *Lemna minor* L. The duck weed caused high algal mortality and probably acted as a physical barrier to ovipositing female mosquitoes.

The present authors investigated the interrelationship of mosquito breeding and four different aquatic environments. The results are reported herein.

**MATERIALS AND METHODS.** This research was conducted during the summer and

fall of 1968 at the Auburn University North Auburn Dairy Research Unit, Lee County, Alabama. Sixteen vinyl plastic wading pools 10 ft. in diameter and 2 ft. deep were assembled in a fenced half acre of sloping pasture adjacent to a small pond. About  $\frac{3}{4}$  cubic yd. of sandy loam topsoil was spread evenly in the bottom of each pool. Unfiltered pond water was pumped into each pool, and appropriate plants were added. The water level was maintained about 6 in. below the rim on the lower side of the pool throughout the research.

The 16 pools were divided randomly into 4 series of treatments with 4 replicates in each series. Each of three series of pools was planted in April and May with one species of aquatic vegetation. The fourth series was not planted. The plants were duck weed, *Spirodela oligorrhiza* (Kuntze) Helgelm, a floating type; elodea, *Egeria densa* Planch., a submerged type; and cattail, *Typha latifolia* L., an emergent type. The pools in the unplanted series developed natural populations of phytoplankton. All plants were able to maintain themselves and grow in size and number. From midsummer to late fall, filamentous algae and extraneous submerged weeds appeared in most of the pools. Grass clippings, leaves, and other debris were often floating on the surface of the pools.

Enough duck weed was placed in each pool to form a complete surface mat which remained in each of the four pools until July 26. At that time about  $\frac{3}{4}$  of the plants were removed from each pool. On September 6 enough duck weed was placed in each of the pools so that the surfaces were covered completely again.

Sampling of immature mosquitoes began June 14 and ended December 12 except for two weeks in September when samples were not taken. The pools were sampled once each week by taking ten dips of water from each pool with a 4.5 in. diam. white enameled dipper. For convenience, it was assumed that every sample was taken randomly, but the only

pools sampled with true randomness were the duck weed series pools. The other pools were sampled primarily in places where mosquito larvae were found most frequently, i.e., around the bases of emergent stems of cattail and in floating algae and debris in elodea and unplanted pools. Both larvae and pupae were collected. The first, second, and third instar larvae were identified to genera. Fourth instar larvae were identified to species. Pupae were identified using larval exuviae or reared adults. No attempt was made to collect adults in the field.

**RESULTS AND DISCUSSION.** Three genera and five species of Culicidae were collected from the simulated natural aquatic environments. These were *Anopheles crucians* Wiedemann, *Anopheles punctipennis* (Say), *Anopheles quadrimaculatus* Say, *Culex erraticus* (Dyer & Knab), and *Uranotaenia sapphirina* (Osten Sacken). The number of larvae of each genus collected in the different habitats are presented in Table 1. *Uranotaenia* larvae were collected only from pools containing emergent vegetation. *Culex* populations exhibited a definite preference for either floating vegetation or emergent vegetation. Generally, they were absent from pools lacking vegetative cover at or above the surface. However, *Anopheles* larvae lived well in floating debris found in elodea pools, in non-planted pools, and around broken leaves and stems in cattail pools. No *Anopheles* larvae were found in the duck weed pools. The plant type

supporting the greatest diversity of mosquito species was emergent vegetation; all five species were collected from pools containing cattails.

The plant association of *Anopheles* spp. based on collections of fourth instar larvae is presented in Table 2. *Anopheles crucians* exhibited a strong preference for emergent vegetation. Apparently, it did not live in floating debris. *Anopheles punctipennis* and *A. quadrimaculatus* occupied all habitats except those containing duck weed.

Disregarding species, vegetation did not significantly enhance the total number of larvae collected from any one habitat. The greatest numbers per dip were collected from habitats containing elodea. Progressively fewer larvae per dip were collected from habitats lacking higher plants, cattails and duck weed, respectively (Table 1). Thus, environments with submerged vegetation produced the largest populations of Culicidae. A Duncan's multiple range test (Table 3) revealed the existence of two significantly different subsets within this population. Although numbers of larvae collected from cattail habitats and unplanted habitats were not significantly different at the 5 percent level, numbers collected from duck weed and elodea habitats were significantly different. This difference would probably have not been significant had only a partial cover of duck weed been maintained throughout the mosquito breeding season.

The most obvious influence of vegeta-

TABLE 1.—Total number of mosquito larvae of each genus collected from each series of plastic pools, Lee Co., Alabama, 1968.

Vegetation	Genus			Total no. collected	Mean number per 10 dips
	<i>Anopheles</i> spp.	<i>Culex erraticus</i>	<i>Uranotaenia sapphirina</i>		
<i>Typha latifolia</i>	235	120	14	369	3.5
<i>Egeria densa</i>	344	2	0	346	4.4**
<i>Spirodela oligorrhiza</i>	0	214*	0	214	2.1
Non-planted	346	28	0	374	3.6

\* All collected during 6-week period when the water surface was not covered by plants.

\*\* Data from one replicate considered atypical and not used.

TABLE 2.—Vegetation preferences of *Anopheles* larvae\* collected from the plastic pools, Lee Co., Alabama, 1968.

Vegetation	Species			Total no. collected
	<i>A. crucians</i>	<i>A. punctipennis</i>	<i>A. quadrimaculatus</i>	
<i>Typha latifolia</i>	8	20	11	39
<i>Egeria densa</i>	0	12	5	17
<i>Spirodela oligorrhiza</i>	0	0	0	0
Non-planted	0	19	18	37
Total	8	51	34	93

\* Fourth instar larvae.

tion on mosquito breeding was by the floating plant, duck weed. Mosquito breeding was completely inhibited when plants were dense enough to cover the pool surface. When this cover was partially removed, breeding produced 0.9 larva per dip. Either larvae were unable to locate a place for their siphons to break the air-water interface or the continuous surface mat presented a barrier to female oviposition. The latter seems a more probable explanation of this inhibitory effect since *Culex* larvae were collected from the pools for 10 days after a continuous layer of plants was replaced.

Habitats containing cattail supported a more consistent and uniform mosquito population. There were three collection peaks between June and November, each producing about 0.8 larva per dip. Elodea and unplanted habitats each yielded no more than 0.5 larva per dip through Au-

gust. Breeding became more intense, however, in early September and in mid-October, and the elodea habitats yielded 2.0 larvae per dip. Unplanted habitats yielded as many as 1.5 larvae per dip. *Anopheles punctipennis* composed the largest part of this population.

## References Cited

- Abdel-Malek, A. 1948. Plant hormones (Auxins) as a factor in the hatching of *Aedes trivittatus* (Coq) eggs. *Ann. Entomol. Soc. Amer.* 41: 51-57.
- Barber, M. A. and Hayne, T. B. 1925. Water hyacinth and breeding of *Anopheles*. *U. S. Public Health Reports* 40:2557-2562.
- Bates, Marston. 1949. *The Natural History of Mosquitoes*. The Macmillan Company, New York, pp. 1-379.
- Bradley, G. H. 1932. Some factors associated with the breeding of *Anopheles* mosquitoes. *J. J. of Agr. Res.* 44(5):381-399.
- Caballero, A. 1919. La Chara faetida A. Br. y las Larvas de *Stegomyia*, *Culex* y *Anopheles*. *Bol. Roy. Soc. Espanola Hist. Nat. Madrid* 19(8):449. Abstract in *Rev. of Appl. Entomol.* 1922B, 10:108.
- Gerhardt, R. W. 1954. Rice fields study report blue-green algae—A possible anti-mosquito measure for rice fields. *Proc. and Pap. of the Calif. Mosq. Contr. Assoc.* 22:50-53.
- Happold, D. C. D. 1965. Mosquito ecology in central Alberta. *Canad. J. Zool.* 43(5):795-819.
- Horsfall, W. R. 1967. Mosquito ecology. *WHO Chronicle* 21(12):525-527.
- Macan, T. T. and Worthington, E. B. 1951. *Life in lakes and rivers*. Collins: London, p. 522.
- MacGregor, M. E. 1924. Tests with *Chara faetida* and *C. hispida* on the development of mosquito larvae. *Parasitology* 16:382-387.
- Rapp, W. F., Jr. and Emil, C. 1965. Mosquito production in an entrophic sewage stabilization lagoon. *J. of Water Pollution Control Federation* 36(6):867-870.

TABLE 3.—Statistical treatment of mosquito collections from each type of vegetation without regard to the species of larvae. Duncan's Multiple Range Test.

Vegetation	Mean number of larvae*
<i>Spirodela oligorrhiza</i>	2.1 a**
<i>Typha latifolia</i>	3.5 ab
Non-planted	3.6 ab
<i>Egeria densa</i>	4.4 b

\* The mean number of larvae per ten dips.

\*\* Means followed by same letter are not significantly different at the 5% level.

- Rueger, M. E., Price, R. D. and Olson, T. H. 1964. Larval habitats of *Culex tarsalis* in Minnesota. Mosq. News 24(1):39-42.
- Smith, J. B. 1910. *Azolla* versus mosquitoes. Entomol. News 21:437-441.
- Smith, W. T., Jr. and Enns, W. R. 1967. Laboratory and field investigations of mosquito populations associated with oxidation lagoons in Missouri. Mosq. News 27(4):462-466.
- Twinn, C. R. 1931. Observations on some aquatic animal and plant enemies of mosquitoes. Canad. Entomol. 63:51-61.
- Zetek, J. 1920. *Anopheles* breeding among water lettuce—A new habitat. Bull. Entomol. Res. 11:73-75.

## THE EFFECTS OF LOW TEMPERATURES ON EGGS OF *Aedes aegypti* (L.)<sup>1</sup>

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### INTRODUCTION

Low winter temperatures are a limiting factor in the survival of many species of insects and, in many instances, restrict their geographical range. An excellent example of such a restricted range is that of *Aedes aegypti* (L.). Christophers (1960) states that the species is probably the only mosquito that, with human assistance, is spread around the globe. He points out that in spite of this wide distribution, it is limited by latitude (45° N and 35° S); distance from the sea; desert conditions; and isolation from human intercourse. Primarily, the northern and southern distributions appear to be related to temperature. There appears to be, with a few exceptions, a striking correlation between the mean isotherm of approximately 50° F for January in the northern hemisphere and July in the southern hemisphere. One of the more notable exceptions is in the eastern portion of the United States where it has been found as far north as Boston, Massachusetts (Christophers, 1960).

Any insect in a cold climate requires

some form of protection against low winter temperatures. This may be the insulating protection of its environment or the ability of the insect to undercool (Salt, 1950). The ability to undercool confers cold-hardiness to an insect species (Salt, 1953, 1961), and many species possess an inherent cold-resistance which enables them to survive extremely low temperatures (Salt, 1956).

This paper reports a portion of the studies conducted by the Technical Development Laboratories (TDL) on the possible inherent cold-resistance in *Ae. aegypti*. Specifically, these experiments were undertaken to determine: (1) if *Ae. aegypti* from different localities (potentially different gene pools) are equally tolerant to low temperatures during the egg stage; (2) if *Ae. aegypti* eggs are capable of surviving temperatures at or near freezing and, if so, for what period of time; and (3) what effect sublethal low temperature exposure has on subsequent larval survival and adult emergence.

### MATERIALS AND METHODS

I. SOURCE OF TEST SPECIMENS. Eggs of strains of *Ae. aegypti* from Puerto Rico; Cucuta, Colombia; Trinidad, B.W.I.; Camp Detrick, Maryland; St. Thomas, Virgin Isles; Pensacola, Florida; Lagos, Nigeria; Sudan; Galveston, Texas; Bangkok, Thailand; Queens, Ontario; and

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