Treehole Productivity for *Aedes triseriatus* in Northern Indiana, 1975 *vs.* 1979

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Introduction

The treehole mosquito, *Aedes triseriatus* (Say), has been known for about eight years as the major vector of LaCrosse Encephalitis in the Midwest (6). LaCrosse, a member of the California group of encephalitis viruses, is responsible for serious illness in hundreds of children in the Midwest; as recent studies show, it may be far more prevalent in human populations than previously thought (3). Therefore, the location and control of this mosquito is important.

Aedes triseriatus breeds primarily in treeholes and rot cavities. However, it has been found increasingly more often in a wide variety of man-made artificial habitats, such as tires, rain gutters and birdbaths. Discarded tires rank as the most common urban source. Several factors make detection and surveillance a difficult endeavor. Conventional surveillance methods for mosquitoes fail to detect significant numbers because *Aedes triseriatus* is not attracted to light or carbon dioxide; moreover, it will not enter live-baited traps (7). The inaccessibility of treeholes has made larval surveys difficult. To date, the best method for detecting the presence of *Aedes triseriatus* has been through the use of a modification of the Fay oviposition trap, in which a blackened aluminum can containing water and a balsawood strip is placed in woodlots to monitor female egg deposition (1).

At present, effective control is impossible. Several control attempts, including the closing of all basal treeholes (2), have resulted in inconsistent results. The inaccessibility of treeholes has led many to suggest biological control measures to reduce population levels. In order to evaluate the effectiveness of any control measures, a more efficient method of surveillance must be found.

Kramer's Woods is an isolated woodlot of roughly 10.1 hectares in size in St. Joseph County, Indiana. The woodlot consists primarily of secondary growth Black and White Oak, creating an abundance of basal treeholes. The surrounding corn fields and pine forests serve to make Kramer's Woods, in effect, an ecologically isolated island in which to study the endemic population of *Aedes triseriatus.*

For these reasons, Dr. Michael Sinsko chose Kramer's Woods to conduct *Aedes triseriatus* population studies in 1975 (5). His survey consisted of locating all the treeholes within the woods and tabulating the size, location, and tree type of each. To monitor the population of *Aedes triseriatus*, he made weekly pupal counts of each treehole from mid-March to mid-September. His method involved emptying the contents of each treehole using a Foley turkey baster, sexing and isolating the pupae into emergence containers, and replacing the contents back into the treehole. He determined both the seasonal production of *Aedes triseriatus* for each treehole and the overall production of the woodlot.

To determine whether or not there is fluctuation in productivity from year to year, or whether the physical makeup of the woods (*i.e.*, number and location of treeholes) changes yearly, a similar study was undertaken in the summer of 1979.

Materials and Methods

The study site, Kramer's Woods, is located 9.6 km northwest of the University of Notre Dame campus. The areas surrounding the woodlot remain relatively unchanged since 1975 with a conifer forest to the north, a cultivated field to the south, and open fields to the east and west, thereby maintaining the isolated character of the woods.

All treeholes were located in early spring following periods of heavy rainfall, using a series of north-south and east-west transect searches. Criteria held for a positive treehole were the ability to hold water and the presence of biological life. Each treehole located was flagged, marked with an aluminum tag, and its size, location, and tree type were recorded (Figure 1).

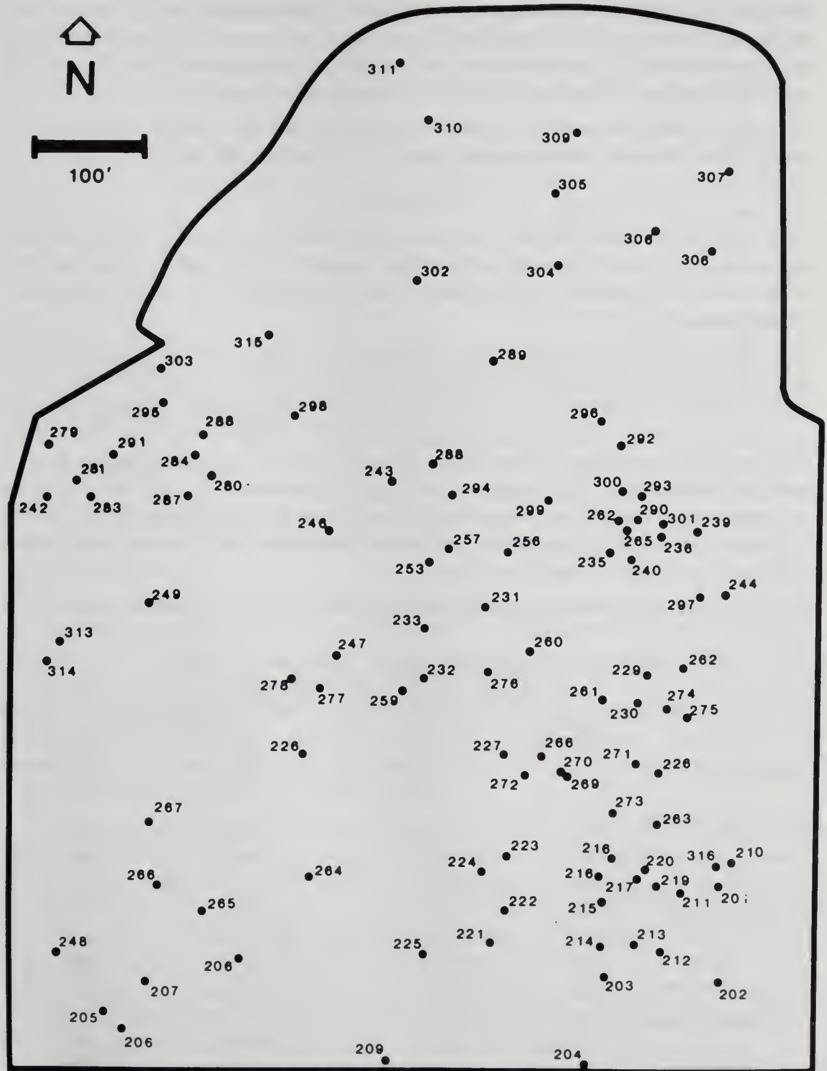

FIGURE 1. *Map of Kramer's Woods, showing treehole distribution in 1979.*

In order to study the *Aedes triseriatus* population of the woodlot, an emergence trap was employed. The emergence traps, modified from Yates (8), were constructed by stapling tubular stockinette (American Hospital Supply) around the hole. A collection vial equipped with an inverted paper funnel at its opening was attached to the distal end of the stockinette with a rubber band. The distal end was secured to a nail or stick so that the collection vial was perpendicular to the treehole.

Emergence traps were set on all treeholes between 900 and 1100 hours, and were collected during the same period the following day. Adults were collected two days per week from June 26 through July 18, three days per week from July 24 through August 31, and two days per week from September 5 through September 20. Adults located in the traps each day were sexed, counted, and released back into the woodlot. It was hoped that this trapping system would yield a representative sample from which total emergence could be estimated.

All rainfall data were obtained from the National Weather Service Office of South Bend, Indiana, Located approximately 2.5 km from Kramer's Woods.

Results

The map of the location of the treeholes found in Kramer's Woods in 1979 shows, as it did in 1975, that the treeholes appear to be clumped throughout the woods with the greatest number being found in the east and southeastern directions (FIGURE 1).

The 1979 survey yielded 104 treeholes, as compared to 108 found in 1975 (TABLE 1). On closer inspection, only 53 treeholes found holding water in 1979 were also recorded in the 1975 survey. This represents a mean yearly treehole turnover of 12.2 trees. The percentage of treeholes in the two types of oak was very close with roughly twice as many Black Oak showing treeholes as White Oak in each of the two years. The examination of the physical parameters of all the treeholes seemed to show a difference between the two years. However, when examined for only those treeholes known to be common to the two surveys, there is very little change in the physical measurements.

The total production of *Aedes triseriatus* was relatively close for each of the

TABLE 1. *Analysis of treeholes in Kramer's Woods, St. Joseph Co., Ind., 1975 vs. 1979*

Treehole character	Year of Analysis	
	1975	1979
Total number	108	104
Species composition in %		
Black Oak	67	63
White Oak	33	37
Dimensions of hole in cm		
All treeholes		
Length	13.0	11.3
Width	8.1	6.9
Area (cm^2)	116.1	87.3
Common treeholes (N = 53)		
Length	12.7	12.1
Width	7.5	7.0
Area (cm^2)	102.2	97.3

Annual rate of treehole turnover = 12.2

TABLE 2. *Treehole productivity for Aedes triseriatus in Kramer's Woods, 1975 vs. 1979*

Adults produced	Year of analysis	
	1975	1979
Female	1790	2207
Male	2438	1393
Total	4228	3600
Sex Ratio, ♀:♂	1:1.4	1:0.63

two years, differing by only 600 individuals (TABLE 2). The difference in the total numbers may be attributed to the relatively low number of males in 1979. There was roughly one-half the number of males in 1979 as there was in 1975, while a slight increase in females was seen in 1979. This is responsible for the reversal of the overall sex ratio from favoring males in 1975 to favoring females in 1979.

Upon examination of the seasonal emergence of both males and females for the two years, there appears to be a striking dissimilarity between the two years (FIGURE 2). However, the general pattern of emergence is the same for each year. In each of the years there is a favoring of males over females in early summer, gradually diminishing to a 1:1 ratio in mid-summer. This is followed by a late summer favoring of females over males. Conspicuously different in the seasonal emergence patterns is the degree to which each of the years favors production of one or another of the sexes in either early or late summer. The high sex ratio favoring males in mid-June of 1975 is not duplicated to such a great extent in 1979. Conversely, there is a greater favoring of females in early September during 1979 than was seen during 1975. During these few weeks in 1979, there were three times more females than males produced within the same period.

The apparent shift in the 1979 emergence is not an environmental event, but rather a reflection of the monitoring method. The 1975 survey utilized pupal counts, while the 1979 survey employed the subsequent adult emergence monitoring techniques, thereby explaining the shifting of the 1979 emergence pattern in relation to 1975.

In an attempt to explain this great difference in the number of *Aedes triseriatus* while the overall seasonal pattern was consistent, the seasonal production was examined in relation to the weekly rainfall. The correspondence between the rainfall and mosquito production shows up very clearly in the 1975 survey (FIGURE 3). The build-up of rain in late May and early June accounts for the peak of pupae produced in mid-June. This is followed by a fluctuating rainfall period that corresponds to moderate pupal production; a period of great rainfall in early August accounts for the late August peaks.

The 1979 data show a moderate amount of rainfall in May, with a build-up in early June accounting for the production of adults in early July (FIGURE 4). This is followed by a relatively dry period with a reduction of emergence. The rapid build-up of rainfall in late July culminates in over 4.2 inches of rainfall during one week in early August. It is during this time that a tremendously high number of treeholes is being filled, with some treeholes beginning to produce larvae for the first that season. This high amount of rain in early August is responsible for the enormous number of adults produced in late August and early September, at a time when a good deal of the treeholes were already dry for the season in 1975.

A ranking of the contribution by each individual treehole to the productivity

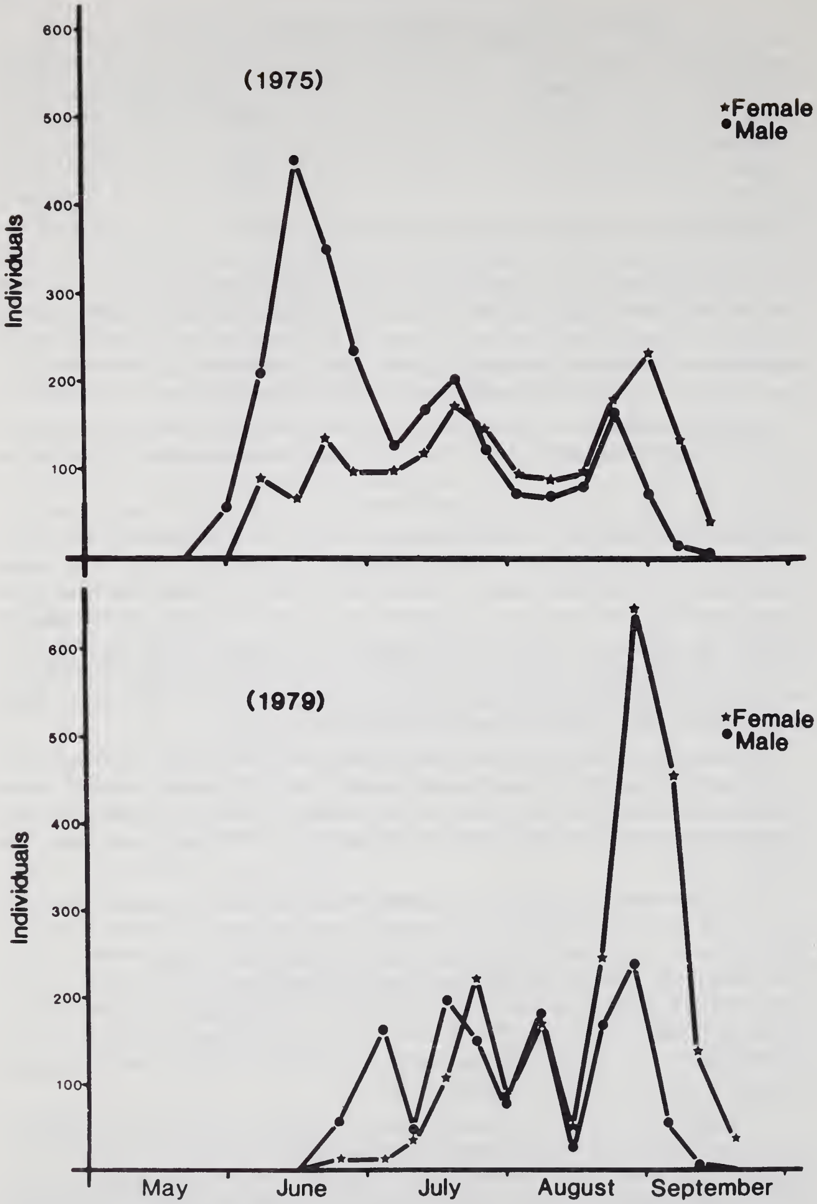

FIGURE 2. *Emergence pattern for each sex of Aedes triseriatus for the years 1975 and 1979.*

of the woodlot (FIGURE 5) reveals a very similar relationship in each year. Of notability is that approximately one-third of the treeholes in each year never produce any *Aedes triseriatus* (37% in 1975 and 35% in 1979).

Sinsko reported that in 1975 over 83% of all pupae were produced by only 22% of the treeholes, with at least two treeholes producing over 500 individuals apiece. A similar event was seen in 1979. However, in 1979, 83% of all adults were produced by 31% of the treeholes, and not more than one treehole approaches even one-half the production of the most productive treeholes in 1975. This illustrates that production of *Aedes triseriatus* was more evenly distributed throughout the treeholes in 1979 than in 1975.

Discussion

The clumped distribution of treeholes in Kramer's Woods may simply be a reflection of the overall distribution of all trees within the woodlot. Sinsko (5), using

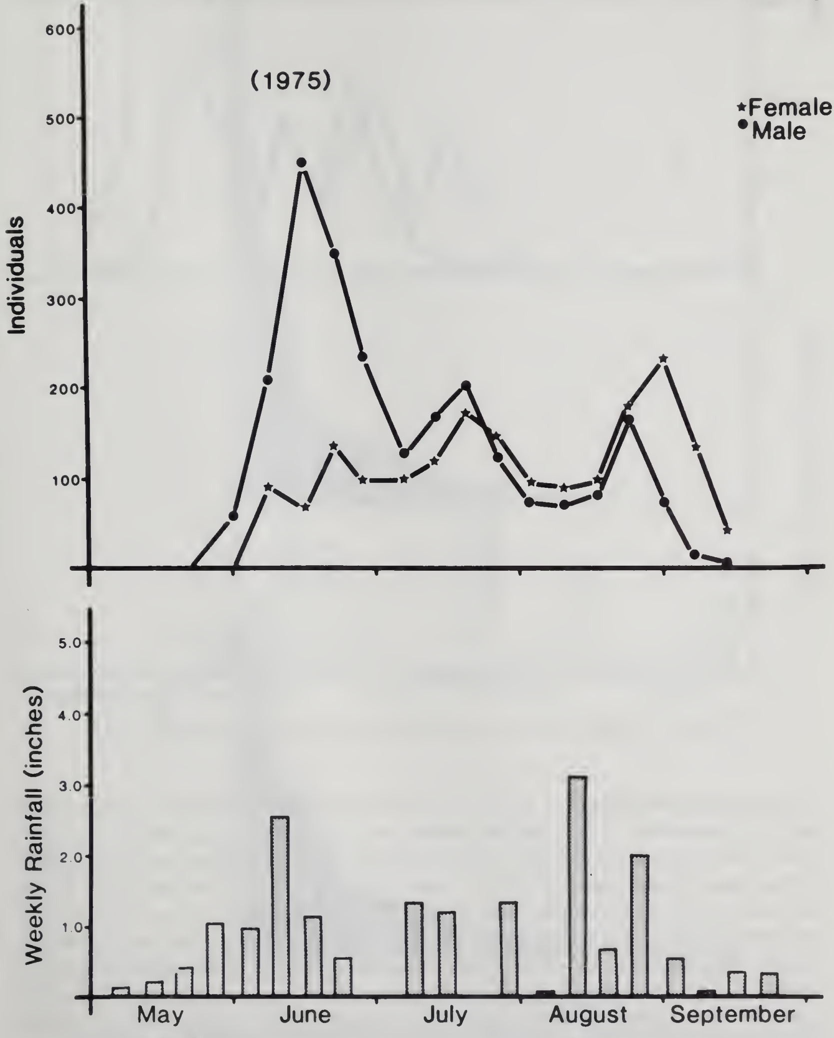

FIGURE 3. *Aedes triseriatus emergence and weekly rainfall for 1975.*

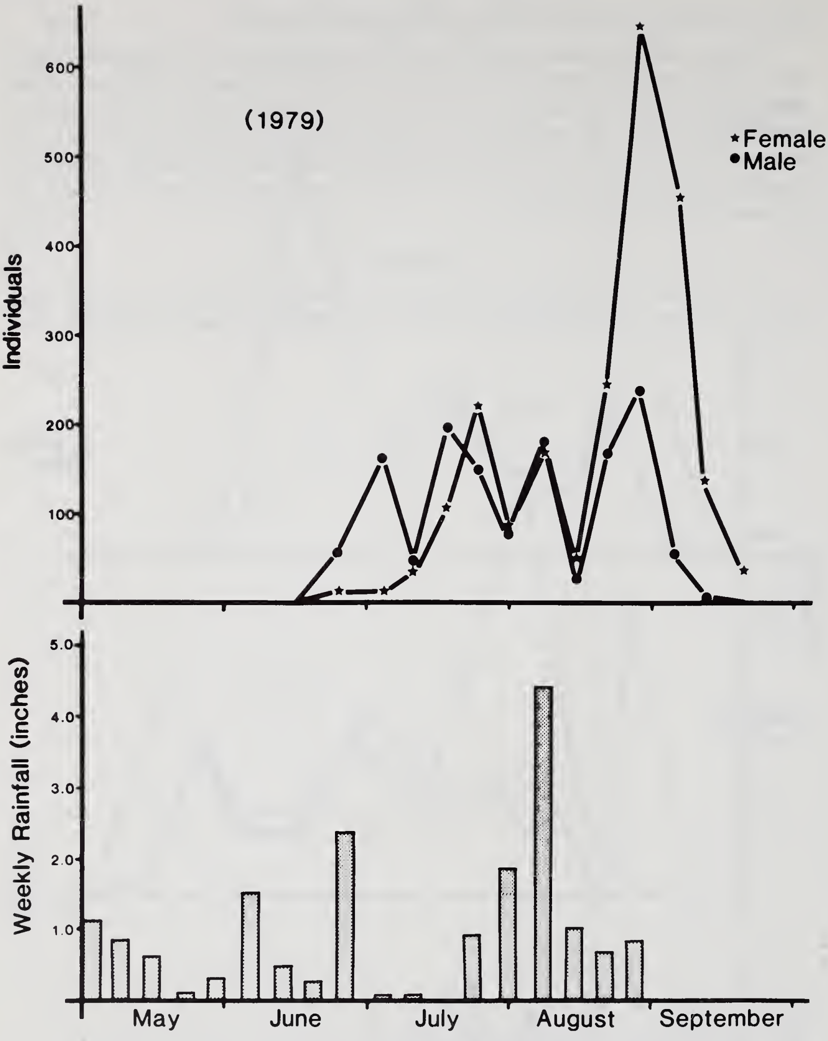

FIGURE 4. *Aedes triseriatus emergence and weekly rainfall for 1979.*

a point-quarter analysis, was able to divide the woods into two distinct area based upon the abundance of saplings and mature trees. His two areas (treehole area and scrub area) correspond roughly to the clumping of the treeholes and the treehole sparse areas. Sinsko calculated that there were 58% Black Oak and 40% White Oak occurring in the treehole area. Therefore, the percentage of each type of oak with treeholes found in either year would not reflect a tendency for one type of oak to form treeholes over the other. The comparison of physical measurements of the treeholes shows that there are distinct populations of treeholes in each of the two years, while the actual treeholes themselves changed very little in size during this period.

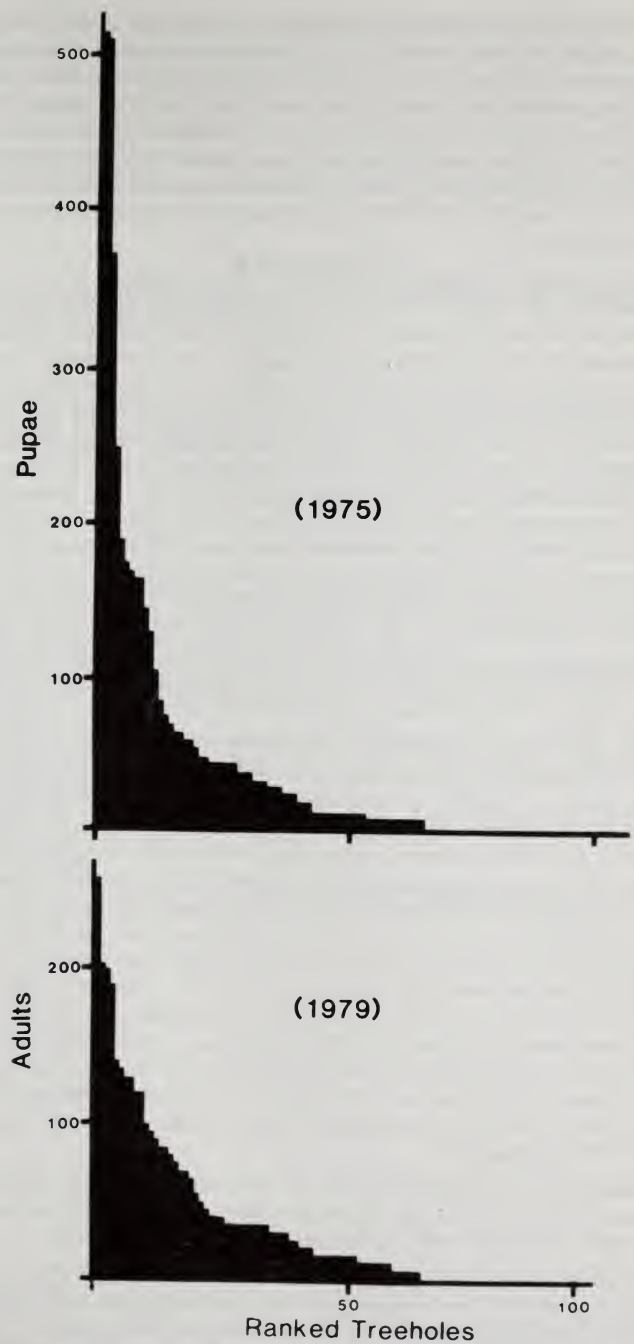

FIGURE 5. *Ranking of treehole productivity for Aedes triseriatus in Kramer's Woods for the years 1975 and 1979.*

The fact that a relatively small number of treeholes endured through both studies, while the total number of treeholes was constant, further demonstrates the fluctuating relationship of the treeholes within the woodlot and the stability of the entire woodlot. This is an excellent example of the difficulties involved in the control of *Aedes triseriatus* based upon the knowledge of the location of all breeding treeholes within a woodlot. As treeholes are added or become no longer capable of holding water each year, control programs based upon location of treeholes would become increasingly difficult without costly searches for new treeholes being done each year.

The seasonal emergence pattern of an early favoring of males gradually switching over to a late season favoring of females in Kramer's Woods has been documented for several years (4,5). Shroyer (4) has suggested several possible mechanisms for the seasonal favoring of one sex or the other based upon a number of environmental conditions and genetic effects. One of his proposed mechanisms states that males seem to exhibit an earlier cessation of overwintering diapause than females, and therefore are seen in much greater numbers early in the summer. Males also tend to have a greater percentage of egg hatching at longer photoperiods (early and mid-summer conditions), with the percentage hatch gradually diminishing as photoperiod shortens (late summer conditions). On the other hand, females exhibit a greater percentage of egg hatching at shorter photoperiods, thereby explaining their overwhelming numbers later in the summer. Shroyer has further stated that, at a certain critical photoperiod, both sexes undergo a dramatic increase in the percentage of eggs that enter into an overwintering diapause, and, therefore, no more hatching of eggs takes place. For Kramer's Woods, Shroyer has calculated the critical photoperiod for females to be around August 10th, with that of males occurring as much as a week earlier.

The examination of emergence patterns for the years 1975 and 1979 support Shroyer's proposed mechanism, as far as the actual production of adults is concerned. In both years, the switchover of emergence from males to females has been exhibited. Of all the years that emergence in Kramer's Woods has been studied, 1979 is the first year that the overall seasonal sex ratio has favored females. This is attributed to the enormous peak of females in early September. This peak of females could be explained, using Shroyer's proposed mechanism, as being due to the great amount of rainfall in early August. If this rainfall occurred before the initiation of the critical photoperiod for females and just after the initiation of the male diapause, it would have had the effect of filling a greater proportion of the treeholes with water, and the hatching out of a number of eggs, the majority of which were females. In August of 1975, there was a good amount of rainfall, but there is not as great a peak of females produced from it. This may be due to the fact that the rains in that year may have been just after the initiation of the critical photoperiod for both sexes, and therefore a good deal of the eggs of each sex had already entered diapause.

Remaining to be explained is the lack of a strong favoring of males in early summer of 1979. It may be that the winter environmental conditions, or the spring temperatures or rains, were not optimal for the hatching of eggs.

The heavy rains in early August of 1979 had the effect of filling a number of previously unproductive treeholes, thereby distributing the production of *Aedes triseriatus* throughout a greater number of treeholes. This is seen in FIGURE 5, in which a greater number of treeholes were needed to produce the same percentage of total mosquitoes as a smaller number of treeholes accomplished in 1975. Of important note here is the observation that a percentage of treeholes in each year never produced *Aedes triseriatus* at all, while a relatively small number of treeholes produced the majority of mosquitoes. This illustrates the importance of better knowledge of the physical, chemical, and environmental parameters of treeholes that attract ovipositing females or that enable larval development, in order to be able to target control measures based upon knowledge of treehole location. In the present study, no correlation was seen between either treehole size or volume and the production of *Aedes triseriatus*.

Acknowledgement

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