A PREFERRED OVIPOSITION SITE AS A SURVEILLANCE METHOD FOR *AEDES AEGYPTI*

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In the current eradication program against *Aedes aegypti* (Schliessmann, 1964) one essential factor for success is a rapid and sensitive surveillance method to evaluate the occurrence of the target species in the field. Since surveillance based on freshly laid eggs would indicate adult females in the field, laboratory studies (e.g., Fay and Perry, 1965) were conducted to determine attractant factors influencing adult females in the selection of an oviposition site and led to the design of a standardized preferred oviposition site for field testing. The present paper compares the results obtained with surveillance based on egg and on larval sampling collections in two urban areas in southern Florida.

PROCEDURE. For the oviposition technique a standard station consisted of a 1-pint, tapered glass jar, painted on the *outside* with a gloss black enamel, and containing a sampling paddle, a vial of ethyl acetate, and approximately 100 ml. of water. The sampling paddle, a wooden tongue depressor to which was glued a panel of brown blotting paper, was held vertically against the inside wall of the jar by masking tape, so that the wet paper panel was exposed and extended from the water to slightly above the upper rim of the jar. The 2-dram homeopathic vial containing A.C.S. grade ethyl acetate was suspended by a wire hook keeping the top of the vial approximately 1 inch below the upper rim of the jar. Where reinspection was at 1 day, 5 ml. of ethyl acetate was put in the vial and, for a 3- or 4-day interval, 10 ml.

The stations were placed in the test areas using one station for every one, two, or four premises, and reinspection was made at 1-, 3- and 4-day intervals. The majority of the stations were placed at ground level in the backyards often adjacent to other potential oviposition sites. Care was taken to select locations having minimum rain drain-off from overhead objects and to avoid locations with prolonged exposure to direct sunlight or high wind currents. Stations numbered to indicate the block and premises were located on a sketch map of the test area.

At reinspection each paddle was checked for eggs which normally were concentrated just above the water line or along

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the edges of the paper panel. A paddle tentatively positive was taped to a carrying board, and a new paddle was placed in the jar. If the station was negative, the original paddle was remounted. Water and ethyl acetate were renewed in each station.

All tentatively positive paddles were checked under a microscope to confirm *A. aegypti* identification since a second species, *Aedes triseriatus,* will oviposit on the paddles.

For the larval sampling technique all premises were inspected. Water in outside artificial containers was carefully checked for *A. aegypti.* Any positive premises were then indicated on a sketch map of the area, and search on those premises was discontinued. Where feasible, all the water was emptied into a dipper for inspection. With larger containers several dip samples were taken from the bottom portions of the water. Water in the leaf axils of certain plants was aspirated with a bulb pipette for examination. A sample of larvae from each infestation was placed in a vial and appropriately labeled for location and type of container. These samples were checked for *A. aegypti* by microscopic examination at headquarters.

Two 5-block areas, each area containing 80 premises, were selected on the basis of potential *A. aegypti* incidence. One area in Perrine, Florida had high levels; the other at South Miami Heights was rated as moderately high. Larval survey of both areas was made by a crew of inspectors August 4–5, 1965. The oviposition survey, made by a single inspector, was initiated on August 10, 1965, with reinspections for positive stations on August 11, 14, and 18, 1965, allowing 1-, 3- and 4-day intervals for egg collections. A second larval survey of both areas was made on August 17–18, 1965, and an estimate of adult occurrence was made at the same time in two blocks. Temperature and rainfall values for the general area were taken for the period July 25 to August 21, 1965.

RESULTS. The results of both types of survey are shown in Table 1. The initial larval survey indicated the assumed estimates of *A. aegypti* incidence to be correct with averages of 12 and 30 percent positive premises in the South Miami Heights and Perrine areas, respectively. It will be noted that the average values for the initial larval and the initial oviposition surveys are quite comparable; however, this is coincidental since the presence of the larvae depends on the collection of rain in artificial containers and the flooding of eggs previously laid. The occurrence of active oviposition would depend on adult females which were pupae on the previous week under minimum time considerations for development.

Further, the data in Table 1 show a marked rise in the number of positive premises from the second oviposition survey to the last oviposition survey. This increase is higher than would be expected merely from the increase of egg-collection time from 3 to 4 days. Reference to the precipitation data for the general area (Table 2), however, offers a plausible explanation for the values obtained.

During the period of July 28–August 1, a total of 2.31 inches of rain was recorded. Rain accumulating in artificial containers would flood and hatch eggs laid in those containers. If July 30 is taken as a median starting point, the larvae in the containers would have been 5 and 6 days old at the time of the first larval survey, making them readily detectable. Average temperatures for the period shown were in the range of 78°–81° F., and development to the pupal stage can be assumed to be 7 to 8 days under these temperatures. The adult female mosquitoes emerged then about August 9–10 and had not reached an egg-laying age at the time of the initial oviposition evaluation on August 11. As oviposition does not normally occur before the females are 4 or 5 days old, the second oviposition survey would reflect oviposition of only the initial brood of these adults. The last oviposition survey, however, covers a 4-day period when all of the adult females, from eggs hatched during the July 28–August 1 period, would

TABLE 1.—*Aedes aegypti* in two test areas as determined by larval and oviposition surveys. Values in parentheses indicate the number of premises sampled

Block Number	Larval Survey 8/4–5 1965	Oviposition Survey 8/10–11 1965	Oviposition Survey 8/11–14 1965	Oviposition Survey 8/14–18 1965	Larval Survey 8/17–18 1965
	South Miami Heights—Positive Premises				
2	2(16)	7(16)	7(16)	15(16)	4(16)
3	2(16)	2(8)	1(8)	7 (8)	0(16)
4	3(16)	0(4)	0(4)	1(4)	1(16)
5	3(16)	1(16)	2(16)	12(16)	2(16)
6	0(16)	0(16)	3(16)	10(16)	0(16)
	South Miami Heights—Percent Positive Premises				
2	12	43	43	93	25
3	12	25	12	87	0
4	19	0	0	25	6
5	19	6	12	75	12
6	0	0	19	62	0
Average	12	15	17	68	9
	Perrine, Florida—Positive Premises				
1	6(16)	7(16)	9(16)	14(16)	6(16)
2	6(16)	1(8)	5(8)	7(8)	5(16)
3	6(16)	2(4)	1(4)	2(4)	0(16)
4	4(16)	0(16)	9(16)	10(16)	4(16)
5	2(15)	8(15)	8(15)	12(15)	3(15)
	Perrine, Florida—Percent Positive Premises				
1	38	43	65	87	38
2	38	12	62	87	31
3	38	50	25	50	0
4	25	0	56	63	25
5	13	53	53	80	20
Average	30	32	52	73	23

TABLE 2.—Rainfall values (inches) for the survey areas July 25 to August 21, 1965.

Date	Inches Rain	Date	Inches Rain	Date	Inches Rain	Date	Inches Rain
7/25	0	8/1	0.34	8/8	0.04	8/15	0
7/26	0.44	8/2	Tr.	8/9	0.11	8/16	0
7/27	Tr.	8/3	0.01	8/10	Tr.	8/17*	0
7/28	0.21	8/4*	0.06	8/11**	0.95	8/18**	0
7/29	0.82	8/5*	0	8/12	0.20	8/19	1.39
7/30	0.15	8/6	0.18	8/13	0.42	8/20	Tr.
7/31	0.79	8/7	0.46	8/14**	0	8/21	0.10

* Date of larval survey.
** Date of oviposition survey.

be ovipositing. Results of the second larval survey would be affected by rainfall during the August 11–13 period which was less than the July 28–August 1 period. Therefore, less eggs would be flooded, resulting in recorded values somewhat lower than those of the initial larval survey.

In this series of surveys, then, the initial larval survey reflects the incidence of developing *A. aegypti* arising from the rainy period of July 28–August 1, and the last oviposition survey reflects the reinfestation of the area by adult females from these larvae. It appears that the larval survey technique is most sensitive 4 to 6 days after a period of heavy rainfall, while the oviposition survey is most sensitive 14 to 18 days after heavy rainfall.

Certain factors of each method of survey must be considered in selecting their suitability for application in field evaluations.

1. *Extent of area surveyed per man day.* Time studies indicate that in a larval survey, one inspector covers approximately two city blocks per day if he inspects each premises within the block. In an oviposition survey one inspector can cover at least six city blocks per day on an initial survey involving placement of a station on each property and revisiting the area for reinspection. For subsequent surveys involving only reinspection one inspector can cover 8 to 10 city blocks per day. In the larval survey the inspector must find and sample all containers on the premises, whereas in the the oviposition survey only one container at most is inspected.
2. *Cost of operation per city block.* Analysis of inspection costs indicate that the oviposition survey can be made for one-half to one-fourth the cost of a larval survey.
3. *Sampling period.* With the larval survey technique the period of highest reliability falls 4 to 6 days following a period of heavy rainfall. During dry seasons eggs may be present in containers but remain undetected as the water level in the containers drops or the water disappears entirely. With the oviposition survey technique highest reliability starts 14 days after heavy rainfall, but egg-laying activity of the females may extend over a 3-week period. The technique gains in reliability during dry periods since a container with water is furnished to the ovipositing females.
4. *Value of results obtained.* The larval technique indicates the areas where *A. aegypti* are developing as the result of eggs hatched by recent flooding but does not indicate the extent of the potential infestation from egg deposition. The oviposition technique conversely indicates the extent of potential infestation but does not indicate the areas where larvae and pupae are developing. Failure to detect freshly laid eggs, larvae, or adults in an area does not necessarily imply eradication since viable but dormant eggs may still exist in dry containers.

With the larval search technique the presence of large larvae or pupae in an insecticide-treated container may indicate insecticide resistance but may also result from inadequate treatment or dissipation of the insecticide. The oviposition technique will not reflect insecticide resistance since the ovipositing females may have originated from untreated larvae or may have migrated from untreated areas.

It is also possible that the oviposition technique can be employed in determining the rate of reinfestation of a cleared area, or in pinpointing isolated foci, but these phases of application will need experimental verification.

CONCLUSIONS. The preliminary field trials comparing the larval survey and the oviposition survey indicate that both methods have utility in field operations. The two techniques are complementary, and the oviposition technique offers an economical, rapid, and sensitive method for determining the presence of adult female *A. aegypti* in the field.

ACKNOWLEDGMENTS. The authors wish to thank Mr. William Prince, Biological Technician, for his excellent performance of the oviposition survey trials in the field and the personnel of the Perrine field station for the larval survey trials. The work described was supported by funds from the Aedes aegypti Eradication Branch of CDC.

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