OVIPOSITION RESPONSE OF AEDES AEGYPTI MOSQUITOES TO DIFFERENT CONCENTRATIONS OF HAY INFUSION IN TRINIDAD, WEST INDIES

DAVE D. CHADEE, ANSON LAKHAN, WAYNE R. RAMDATH AND ROBIN C. PERSAD

Insect Vector Control Division, Ministry of Health 3 Queen Street, St. Joseph, Trinidad, West Indies

ABSTRACT. Ovitraps containing various concentrations of hay infusion and tap water were exposed weekly in the field for 15 wk to determine the oviposition patterns of *Aedes aegypti*. The results showed 10, 20, 60 and 80% hay infusions each attracted similar numbers of *Ae. aegypti* eggs oviposited and egg occurrences. No repellent effect was observed. In another field study, significantly more eggs and egg occurrences were collected from 25 and 50% hay infusions and tap water. The differences in these results from those of a previous study in Puerto Rico are discussed.

Numerous workers have demonstrated that decomposing organic matter associated with water is attractive to *Aedes aegypti* (Linn.) (Buxton and Hopkins 1927, O'Gower 1963). Gjullen et al. (1965) reported that grass infusion and log pond water as compared to distilled water increased oviposition by *Ae. aegypti* and *Culex quinquefasciatus* Say. However, Hazard et al. (1967) showed that *Ae. aegypti* was not attracted to hay infusion odor in an olfactometer but was stimulated to oviposit by an unknown compound in these solutions.

Reiter et al. (1991) utilized 2 ovitraps, one ovitrap contained 100% hay infusion and an adjacent ovitrap that contained a 10% dilution of hay infusion in tap water to attract gravid females. However, it is uncertain whether the 100% hay infusion serves as a mosquito attractant because with increasing concentrations, the activity of the infusion can change from an attractant to a repellent (Dethier 1947).

This study investigated field oviposition patterns of *Ae. aegypti* populations exposed to various concentrations of hay infusions in St. Joseph, Trinidad, from June 17 to September 26, 1991, a period within the rainy season (May-November). The study area, meteorology and population of *Ae. aegypti* have been described by Chadee and Corbet (1987). The hay infusion was prepared weekly in a similar manner to Reiter et al. (1991) except that the dried hay grass was a local species, *Sclerica bractaeda* Linn.

Field trial I: Nine dilutions were made from the broth infusion (100%). The dilutions were made with tap water in 10% decrements (i.e., 90, 80, 70%, etc.) and a tap water control for each field experiment. In the field, 5 replicates of 10 attractant concentrations and a control were used. Oviposition of *Ae. aegypti* was monitored using ovitraps (Fay and Eliason 1966) as described by Chadee and Corbet (1987) and were placed under the western eaves of 5 houses to prevent rainfall from further diluting the broth concentrations. At each site, 11 ovitraps were each allotted 350 ml of the appropriate attractant concentration. These ovitraps were located linearly 5 cm from nearest neighbor under the eaves. The broth was transported in sealed containers and poured into ovitraps in the field. Ovitrap "paddles" made of brown hardboard cut into thin strips (12.5×2.5) cm) onto which mosquitoes lay eggs just above the water level, were numbered in accordance with the hay infusion treatment and ovitraps were set out between 1000 and 1100 h, the time of lowest oviposition activity (Chadee and Corbet 1987). These trials were conducted on the same day each week. Ovitraps were exposed for 1 day each week for 15 wk. After each 24-h exposure period, paddles were removed, the media in each ovitrap was discarded, and the ovitrap was scrubbed to remove any eggs attached to the insides of the ovitrap and returned to the laboratory. Postcollection handling of paddles and identification of eggs have been described by Chadee and Corbet (1987).

Field trial II: A companion experiment using 100, 75, 50, 25 and 10% broth infusion and a tap water control was set up in another study area within 1 km of St. Joseph. All procedures were in accordance with those described in field trial I.

During both studies, oviposition activity as recorded as occurrence of eggs and as the number laid per week. Field data were analyzed using a G-test and Kendell coefficient of rank correlation test (Sokal and Rohlf 1980) to determine oviposition response to different treatments.

The results of the 1st field trial are shown in Table 1. All ovitraps exposed were used by gravid *Ae. aegypti* females. The distribution of eggs among the 55 ovitraps appears to be random (P = 0.15) because no significant relationship can be detected between the number of eggs laid and position or distribution of ovitraps used along

Ξ

Table 1. Occurrence and number of Ae. aegypti eggs collected from 5 replicates of 11 ovitraps exposed for 15 wk.

Ovi-	Contents Eggs collected			
trap no.	of ovi- trap ¹	Mean ± SD	Total	Occur- rence
1	Water	$\frac{-52}{18.5 \pm 26.6}$	271	15
2	10% HI	67.6 ± 52.2	1.071	29
3	20% HI	47.4 ± 45.6	772	25
4	30% HI	32.5 ± 40.2	506	20
5	40% HI	18.7 ± 24.2	364	12
6	50% HI	28.6 ± 44.9	493	16
7	60% HI	52.8 ± 73.9	781	16
8	70% HI	28.4 ± 49.6	297	16
9	80% HI	57.8 ± 61.5	869	21
10	90% HI	26.7 ± 39.6	453	12
11	100% HI	45.5 ± 48.4	680	17
Total		33.0 ± 48.4	6,557	199

' HI = hay infusion.

the 5 sites (Kendell coefficient of rank correlation test [Sokal and Rohlf 1980] used in both cases). In addition, no single concentration of hay infusion was significantly more attractive than any other and no concentration showed a repellent effect (271 eggs in tap water vs. 297 in 70% hay infusion) (Table 1). Large numbers of *Ae. aegypti* eggs and oviposition occurrences were recorded from 10% (1,071 eggs), 20% (772 eggs), 60% (781 eggs) and 80% (869 eggs) hay infusion, but these egg numbers and occurrences were not significantly different (G = 1.45, df = 10, NS). Thus, it can be assumed that *Ae. aegypti* mosquitoes were similarly attracted to these hay infusion concentrations.

Data from the 2nd field trial are presented in Table 2. There were no significant differences in the oviposition responses of *Aedes aegypti* to different concentrations of hay infusion (G = 2.0, df = 5, NS). These results indicate that similar numbers of eggs and oviposition occurrences were collected at 25% (275 eggs) and 50% (254 eggs) hay infusion as well as in the control (274 eggs) (Table 2).

It is clear that tap water and various concentrations of hay infusions are all more or less equally attractive to gravid *Ae. aegypti* mosquitoes. The fact that tap water and 10, 25 and 50% hay infusions were attractive during both field trials indicates that there is no significant difference in preferences. The wide range of hay infusion concentrations used by *Ae. aegypti* during the present field trials may help explain the wide range of aquatic habitats used by gravid *Ae. aegypti*; that is from clean water (Christophers 1960) to water in septic tanks (Babu et al. 1983).

Table 2. Occurrence and number of *Ae. aegypti* eggs collected from 6 concentrations of hay infusion (HI) in Trinidad, West Indies (1991).

Concen-	Eggs colle	Occur-	
tration ¹	Mean ± SD	Total	rence
0 (water)	45.7 ± 28.3	274	6
10% HI	37.3 ± 39.1	149	4
25% HI	55.0 ± 40.0	275	5
50% HI	50.8 ± 41.8	254	5
75% HI	27.8 ± 32.3	111	4
100% HI	28.3 ± 16.9	113	4
Total	40.8 ± 33.0	1,176	28

¹ HI = hay infusion.

Hazard et al. (1967) demonstrated the presence of an unidentified *Ae. aegypti* arrestant and/ or oviposition attractant isolated from hay infusion. Further studies showed that *Ae. aegypti* was not attracted to the odors of hay infusion in an olfactometer though the mosquitoes were stimulated to oviposit in these hay infusion solutions. If *Ae. aegypti* was not attracted to the odor of hay infusion (Hazard et al. 1967), then using "enhanced pairs" as suggested by Reiter et al. (1991) may be inappropriate.

The differences found between the present results and those of Reiter et al. (1991) further demonstrate the variability in oviposition response. In addition, this variability may reflect the differences in the type of hay used to make the infusion, differences in methodology (2 concentrations compared vs. 11 concentrations) and the difficulty in the standardization of hay infusion from one country to another.

We thank N. Boodoosingh, R. Ganesh and G. Bally for field and laboratory assistance. Special thanks are due to J. M. Sutherland for reviewing the manuscript.

REFERENCES CITED

- Babu, C. J., K. N. Panicker and P. K. Das. 1983. Breeding of *Aedes aegypti* in closed septic tanks. Indian J. Med. Res. 77:637.
- Buxton, P. A. and G. H. E. Hopkins. 1927. Researches in Polynesia and Melanesia. Parts I-IV. Mem. Lond. School. Hyg. Trop. Med. 1:1-260.
- Chadee, D. D. and P. S. Corbet. 1987. Seasonal incidence and diel patterns of oviposition in the field of the mosquito, *Aedes aegypti* (L.) (Diptera: Culicidae) in Trinidad, W.I.: a preliminary study. Ann. Trop. Med. Parasitol. 81:151-161.
- Christophers, S. R. 1960. Aedes aegypti (L.). The yellow fever mosquito. Cambridge Univ. Press, Cambridge.
- Dethier, V. G. 1947. Chemical insect attractants and repellants. Blakiston, Philadelphia.

- Fay, R. W. and D. A. Eliason. 1966. A preferred oviposition site as a surveillance method for Aedes aegypti. Mosq. News 26:531-535.
- Gjullin, C. M., J. O. Johnsen and F. W. Plapp, Jr. 1965. The effect of odors released by various waters on the oviposition sites selected by two species of Culex. Mosq. News 25:268-271.
- Hazard, E. I., M. S. Mayer and K. E. Savage. 1967. Attraction and oviposition stimulation of gravid female mosquitoes by bacteria isolated from hay infusion. Mosq. News 27:133-136.
- O'Gower, A. K. 1963. Environmental stimuli and the oviposition behavior of Aedes aegypti var. queenslandis Theobald (Diptera: Culicidae). Anim. Behav. 11:189-197.
- Reiter, P., M. A. Amador and N. Colon. 1991. Enhancement of the CDC ovitrap with hay infusion for daily monitoring of Aedes aegypti populations. J. Am. Mosq. Control Assoc. 7:52–55. Sokal, R. R. and F. J. Rohlf. 1980. Biometry, 2nd
- ed. Freeman, New York.