

INFLUENCE OF VEGETATION ON THE SPATIAL DISTRIBUTION OF *TOXORHYNCHITES MOCTEZUMA* OVIPOSITIONS IN THE FIELD

SIMON JORDAN¹ AND STEPHEN F. HUBBARD

Department of Biological Sciences, University of Dundee, Dundee, Scotland DD1 4HN, U.K.

ABSTRACT. The influence of the vegetation surrounding black-painted calabash ovitraps on the number of eggs of the mosquito *Toxorhynchites moctezuma* they attracted was investigated using oviposition data gathered from a seasonal-deciduous forest in Trinidad, West Indies. More eggs were laid into ovitraps situated either within or directly adjacent to trees or bamboo stools than those not associated with trees or bamboo. This result is discussed in terms of the initial oviposition-site searching behavior of female *Toxorhynchites*.

Mosquitoes of the genus *Toxorhynchites* (Theobald) are non-hematophagous, and as such present no medical threat to humans. Indeed, because the aquatic larval stages of all *Toxorhynchites* species are predacious upon other small aquatic arthropods, including other mosquito larvae that share their container environment, they represent potential biological control agents of mosquito pests and disease vectors. In certain situations (Gerberg and Visser 1978, Bailey et al. 1983) they have been shown to be an effective means of suppressing vector populations.

In recent years, much effort has been directed toward understanding the oviposition-site preferences of various species of *Toxorhynchites* (Trimble 1979; Durso et al. 1982; Focks et al. 1983a, 1983b; Hilburn et al. 1983; Benzon et al. 1988; Linley 1988; O'Malley et al. 1989). This is not surprising, because a major impediment to their use as biological control agents seems to be the inability of released females to find and oviposit in the same containers as the target species. The reasons for this are unclear, but in *Toxorhynchites rutilus rutilus* (Coq.) at least, there appears to be a very strong vegetational influence on oviposition-site selection. When 160 male and 160 female *Tx. r. rutilus* were released into a 9-block residential area of New Orleans, this species was so oriented to trees that ovipositions were recorded from less than about 20% of the available artificial oviposition sites (Focks et al. 1983a). An ovitrap was likely to attract ca. 40% more ovipositions if it was associated with trees and vegetation than if situated, for example, at the base of a house in an open yard. A similar release of *Toxorhynchites amboinensis* (Doleschall) in the same neighborhood revealed this species to be much more catholic in its preferences, and willing to oviposit into almost any kind of artificial container,

including those rejected or otherwise overlooked by *Tx. r. rutilus* (Focks et al. 1983b). Similarly, Bailey et al. (1983) showed that a dense natural population of *Tx. r. rutilus* significantly reduced a natural population of *Aedes aegypti* (Linn.) in a tire dump located under trees, but had little effect in one in the open, despite releases of 1,000 adult *Toxorhynchites* per week. They attributed this result to an ovipositional preference for shade afforded by the trees, rather than the trees themselves.

Although it is clear that females of *Tx. r. rutilus* are attracted toward a tree or bush in an otherwise open yard (Focks et al. 1983a) and to wooded plots rather than open ones (Bailey et al. 1983), it is not known if females of other species will also display such preferences or if they would be drawn preferentially to ovitraps adjacent to trees in a densely forested environment. In these notes, we report the analysis of data which indicates the existence of such vegetational influences on the oviposition of *Toxorhynchites moctezuma* (Dyar and Knab) in tropical rainforest in Trinidad, West Indies.

The study was conducted during June, July and August of 1987 at the Simla Research Station (10° 42' N, 61° 17' W), located on the eastern side of the Arima Valley in the Northern Mountain Range at an elevation of 250 m, about 10 km north of the town of Arima, Trinidad, West Indies.

The original vegetation of the study area is comprised of the *Inga macrophylla* H. & B. ex Willd. *Guarea quara* (Jacq.) P. Wilson (pois doux-redwood) association characteristic of lower montane deciduous forest.

In the past this vegetation has been cleared or otherwise much disturbed by cultivation, quarrying or the removal of valuable hardwoods. In addition there are many plantation species including cocoa, banana and citrus, and many stands of bamboo, all of which contribute to a secondary seasonal-deciduous association. A more thorough description of the vegetation of the Arima Valley is given by Beebe (1952) and

¹ Current address: Florida Medical Entomology Laboratory, 200 9th Street S.E., Vero Beach, FL 32962.

of the research station area by Smith et al. (1986).

Ovitrap of the "black-painted calabash" design were prepared by cutting a hole of approximately 12–13 cm diam in the fruits of *Crescentia cujete* (Linn.) Bignoneaceae, scooping out the fleshy pulp within, drying the thick, woody epicarps in the sun and painting them with matt-black paint (Hubbard, Chadee and O'Malley, unpublished data). One hundred seventy-five such ovitraps were then set out in the forests surrounding the research station as 7 grids, each comprising 25 ovitraps spaced 5 m apart in a 20-m square. These grids were situated at randomly selected sites between 50–450 m from the research station. Ovitrap were placed on the ground with the open end facing about 45° to the horizontal, and were originally filled with between 250–2,220 ml of spring water (depending on the size of calabash ovitrap), thereafter being topped up by rainfall.

Each day for 84 consecutive days, every ovitrap was inspected, and all *Toxorhynchites* eggs removed from the water surface using a modified plastic spoon (Chadee and Small 1988) and recorded. The ovitraps were visited in the same order each day, starting at 0800 h and finishing between 1000 and 1100 h, depending on the number of eggs harvested. Visits were made at these times because Chadee et al. (1987) found no oviposition before noon, so sampling in this way was less likely to disturb ovipositing *Tx. moctezuma* females. All eggs were brought back to the laboratory in water-filled vials, labeled with date and site of oviposition, reared through to the adult stage and identified as *Tx. moctezuma*.

The original objective of this experimental design was to gather data on the seasonal pattern of oviposition, however, after preliminary analysis of the data, significant differences between grids in the mean number of ovipositions received per trap per day were detected. The

physical proportions of the traps themselves did not yield any clue as to why such a phenomenon should occur, but it was noticed that the grid with the highest mean number of eggs per trap per day had distinctly different vegetation than any other grid in that it had more bamboo. Therefore, to investigate the possibility that vegetation was a factor affecting oviposition, the physical vegetative characteristics of the microhabitat of each ovitrap, defined as a circle of 1 m diam around each ovitrap, were categorized into one of 7 "vegetation types." These were: 1) open—shaded by the canopy yet devoid of vegetation; 2) sparse vegetation—shaded by canopy supporting 10 or less scattered shrubs and saplings; 3) dense vegetation—shaded by canopy, supporting more than 10 shrubs and saplings; 4) in the crotch of the buttressed roots of a tree; 5) adjacent to a tree trunk (no buttressed roots); 6) adjacent to a stool of bamboo; and 7) within a stool of bamboo.

Ovitrap in the crotches of buttressed roots and within stools of bamboo achieved the highest indices of attractivity (Focks et al. 1983a) at 1.8 and 1.7%, respectively (Table 1). The least attractive traps were those in the "open" category of vegetation, which received only 0.2% of the total oviposition per trap. Clearly some vegetation types or property associated with them were much more attractive to *Tx. moctezuma* than others, but to confirm the statistical significance of this, a one-way analysis of variance was performed on the data representing the total numbers of eggs recovered from each ovitrap. This revealed highly significant differences between the vegetation types ($F = 17.08$, $P < 0.001$).

These results, although obtained through an unplanned *a posteriori* test, seem to indicate that oviposition by *Tx. moctezuma* into calabash ovitraps was strongly influenced by the vegetation immediately surrounding them and was greatly enhanced when ovitraps were in close proximity

Table 1. Distribution of *Toxorhynchites moctezuma* ovipositions in tropical rainforest in Trinidad, W.I. (1987).

Vegetation type	No. of ovitraps	No. of eggs recovered	% of total	Mean no. eggs per ovitrap (\pm S E)	% of total oviposition per ovitrap
Open	24	317	5.5	13.2 \pm 2.3	0.2
Sparse	65	1,496	26.1	23.0 \pm 2.1	0.4
Dense	41	1,004	17.5	24.5 \pm 2.5	0.4
In crotches	9	906	15.8	100.7 \pm 29.7	1.8
By tree	18	949	16.8	52.7 \pm 8.1	0.9
By bamboo	11	680	11.9	61.8 \pm 14.4	1.1
In bamboo	4	380	6.6	95.0 \pm 29.5	1.7
Total	172*	5,732	100.0		

* Three of the original 175 ovitraps were destroyed by falling trees and not replaced, therefore they were omitted from the analysis.

to trees and bamboo. This may be expected, because it is within trees and bamboo that their natural oviposition sites occur. It is less certain, however, why other ovitrap sites appeared not to attract as many ovipositions—the traps were readily available to searching females, and were of an identical design to those associated with trees or bamboo; only the vegetation surrounding them was different. There are 2 possible explanations for the apparent low attractivity of these traps; either females are rejecting traps as unsuitable due to the vegetation around them, or they are failing to locate them in the first place.

In the laboratory, several authors have identified factors that strongly influence the ovipositional decisions made by female *Toxorhynchites*. The color of the site (Trimble 1979), presence of water (Benzon et al. 1988) and water chemistry (Benzon et al. 1988, Linley 1988) have all been shown to be important cues that the female must consider before committing her progeny to their preadult environment. However, these are internal properties of the ovitrap itself, not external factors such as vegetation. It seems unlikely that a female *Toxorhynchites* would reject a suitable oviposition site, once she had located it, on the basis of the type of vegetation around it. A more plausible answer for the pattern of oviposition described above is that traps not associated with trees or bamboo have a reduced chance of being located by female *Toxorhynchites*, possibly because of the females' search behavior. Based on the observed distribution of eggs in artificial containers after an urban adult release, Focks et al. (1983a) suggested that *Tx. r. rutilus* females first orient themselves in close proximity to places where oviposition sites occur most frequently, i.e., trees. Our analysis suggests that this is also the case with *Tx. moctezuma*, and that bamboo stools seem to be one of the primary cues too. Once such an orientation has been achieved, females presumably search visually for dark crevices and holes, and once these have been located, utilize their hygro- and chemosensory apparatus to make a final decision whether to oviposit. Such a sequence of behavioral events has yet to be observed in the field, but it would certainly be an efficient way of finding oviposition sites. It would also explain why the 42 ovitraps associated with trees or bamboo (24.4% of the total number of ovitraps) accounted for more than 50% of the total number of eggs collected.

We thank A. D. Gettman, J. R. Linley and L. P. Lounibos for their critical reviews of an earlier draft of the manuscript. Facilities were provided by Deosaran Sinanan, Jonathan Stuart and the Asa Wright Nature Centre. Financial

assistance was provided by the Natural Environment Research Council (S.J.) and the sponsors of the Dundee University Rainforest Expedition 1987 to whom we are extremely grateful.

REFERENCES CITED

- Bailey, D. L., R. G. Jones and P. R. Simmonds. 1983. Effects of indigenous *Toxorhynchites rutilus rutilus* on *Aedes aegypti* breeding in tire dumps. Mosq. News 43:33-37.
- Beebe, W. 1952. Introduction to the ecology of the Arima Valley, Trinidad, B.W.I. Zoologica 37:157-183.
- Benzon, G. L., C. S. Apperson and W. Clay. 1988. Factors affecting oviposition site preference by *Toxorhynchites splendens* in the laboratory. J. Am. Mosq. Control Assoc. 4:20-22.
- Chadee, D. D., S. F. Hubbard and P. S. Corbet. 1987. Diel patterns of oviposition in the field of *Toxorhynchites moctezuma* (Diptera: Culicidae) in Trinidad, West Indies. J. Med. Entomol. 24:1-5.
- Chadee, D. D. and G. J. Small. 1988. A simple spoon device for collecting eggs of *Toxorhynchites* in the laboratory and field. J. Fla. Anti-Mosq. Assoc. 59:5-6.
- Durso, S. L., J. D. Demaio and J. C. Beier. 1982. Ovipositional behavior of *Toxorhynchitesamboinensis* in a tire yard. Mosq. News 42:255-260.
- Focks, D. A., S. R. Sackett, D. A. Dame and D. L. Bailey. 1983a. *Toxorhynchites rutilus rutilus* (Diptera: Culicidae): field studies on dispersal and oviposition in the context of the biocontrol of urban container-breeding mosquitoes. J. Med. Entomol. 20:383-390.
- Focks, D. A., S. R. Sackett, D. A. Dame and D. L. Bailey. 1983b. Ability of *Toxorhynchitesamboinensis* (Doleschall) (Diptera: Culicidae) to locate and oviposit in artificial containers in an urban environment. Environ. Entomol. 12:1073-1077.
- Gerberg, E. J. and W. M. Visser. 1978. Preliminary field trial for the biological control of *Aedes aegypti* by means of *Toxorhynchitesbrevipalpis*, a predatory mosquito larva. Mosq. News 38:197-200.
- Hilburn, L. R., N. L. Willis and J. A. Seawright. 1983. Analysis of preference in the color of oviposition sites exhibited by female *Toxorhynchites r. rutilus* in the laboratory. Mosq. News 43:302-306.
- Linley, J. R. 1988. Laboratory experiments on factors affecting oviposition site selection in *Toxorhynchitesamboinensis* (Diptera: Culicidae), with a report on the occurrence of egg cannibalism. Med. Vet. Entomol. 2:271-277.
- O'Malley, S. L. C., S. F. Hubbard and D. D. Chadee. 1989. Oviposition habitat preferences of *Toxorhynchites moctezuma* mosquitoes in four types of tropical forest in Trinidad. Med. Vet. Entomol. 3:247-252.
- Smith, J. A. C., H. Griffiths and U. Lutge. 1986. Comparative ecophysiology of CAM and C3 bromeliads. I. The ecology of the Bromeliaceae in Trinidad. Plant. Cell Environ. 9:359-376.
- Trimble, R. M. 1979. Laboratory observations on oviposition by the predaceous tree-hole mosquito, *Toxorhynchites rutilus septentrionalis* (Diptera: Culicidae). Can. J. Zool. 57:1104-1108.