District's mosquito control efforts have increased as a result.

An indirect benefit derived by the District has occurred as a by-product of the system analysis that was required to implement the computer system. The kinds of data required and the logic used in decision making were reviewed, re-evaluated and modified as required. The result has been a clarification of information flow and decision making procedures.

References Cited

Roberts, F. C. and G. E. Conner. 1979. Use of a mathematical model to predict levels of adult *Culex pipiens* in Alameda County. Proc. Calif. Mosq. Vector Control Assoc. 47:114–115.

Schooley, J., F. C. Roberts and G. E. Conner. 1982. A preliminary simulation of mosquito control in Coyote Hills freshwater marsh, Alameda County, California. Proc. Calif. Mosq. Vector Control Assoc. 50:76–81.

GONOTROPHIC STATE AND PARITY OF NECTAR-FEEDING MOSQUITOES

AGNES M. VARGO AND WOODBRIDGE A. FOSTER

Department of Entomology, Ohio State University, 1735 Neil Avenue, Columbus, OH 43210

ABSTRACT. Mosquitoes were collected on autumn evenings in Ohio while they were feeding on snakeroot and goldenrod flower nectar. Females of *Aedes vexans* and *Culex restuans* were sufficiently numerous to indicate when, in the gonotrophic cycle and in the adult life span, they typically took nectar. Dissections of 444 females indicated that both species seldom fed on nectar while digesting a blood meal (3–4% were blooded). The great majority of *Ae. vexans* took nectar while empty (79%) (neither blooded nor gravid), whereas *Cx. restuans* took nectar as commonly when gravid (49%) as when empty (48%). Parous nectar feeders were common among both species. A comparison of the parous/nulliparous ratio of nectar-feeding and blood-seeking collections of *Ae. vexans* indicated that nectar feeding was most frequent among nulliparous females but continued throughout adulthood.

INTRODUCTION

Many observations have been recorded of female mosquitoes feeding on plant nectars (Hocking 1953), and the utilization of nectar sugars for dispersal, reproduction, and survival has been demonstrated (Nayar and Sauerman 1971, 1975a, 1975b, 1975c; Nayar and Van Handel 1971). Sugar feeding appears to inhibit blood-feeding behavior temporarily in *Aedes aegypti* (Linn.) (Jones and Madhukar 1976) and to delay oviposition in several species (see Shroyer and Sanders 1977). Since both longevity and biting frequency are important to a mosquito's ability to transmit pathogens, knowledge of the details of the timing of nectar feeding may prove useful in epidemiological studies. In the present study we examined the gonotrophic state and reproductive age (parity) of mosquitoes in central Ohio in order to determine when, in the gonotrophic cycle, nectar feeding is most likely to occur, and whether it is a repetitive event.

Most reports of female nectar feeding have not noted whether any of the mosquitoes were digesting blood or were gravid at the time. Philip (1943), de Meillon et al. (1967) and Brantjes and Leemans (1976) indicated that nectar-feeding mosquitoes frequently contain blood meals or mature eggs, but the data of McCrae et al. (1976) and Magnarelli (1977, 1978) suggest that mosquitoes are usually empty of both blood and eggs when they take nectar. Magnarelli (1978, 1980) has provided indirect evidence that mosquitoes take nectar repeatedly throughout adulthood; resting and blood-seeking females contained fructose about as frequently when they were parous as when they were nulliparous. Direct comparisons of the parous/nulliparous ratios of nectar-feeding mosquitoes and the mosquito population at large apparently have not been published.

MATERIALS AND METHODS

Collections were made in a narrow wooded area between Union Cemetery and the Olentangy River in Columbus, Ohio, on 13 days from September 16 to October 19, 1979. Sampling began 0.5 hr after sunset and continued for approximately 2 hr or until mosquito ac-

tivity ended. With the aid of a head lamp, mosquitoes were collected with a mouth aspirator while they were either probing flowers or holding the proboscis in nectaries of flowers of white snakeroot, *Eupatorium rugosum* Houtt, and Canada goldenrod, *Solidago canadensis* Linn. Blood-seeking mosquitoes were collected simultaneously from the exposed arms of the collectors to provide a sample of the age structure of the mosquito populations in the area. All mosquitoes were held in screened cartons and frozen at −40°C within 1 hr after the collection period.

Later, females were identified to species and classified according to gonotrophic state: empty (no blood in gut, ovarian follicles undeveloped), blooded (blood in gut), or gravid (no blood in gut, eggs mature). Blooded mosquitoes were subdivided according to the scheme of W.H.O. (1975) into freshly fed, late fed, half-gravid, and subgravid. Ovaries of empty females were removed in distilled water and categorized as nulliparous or parous according to the tracheolar skein method (Detinova 1962). Parity could not be determined in a few cases when ovaries were damaged. To establish whether apparently freshly-fed blood-seeking mosquitoes had surreptitiously taken a blood meal from the collector before being caught, their follicles were classified according to Christophers (1911) as modified by Mer (1936). If follicles had developed beyond late stage II, it was concluded that they were seeking a supplementary blood meal.

A chi-square test for "goodness-of-fit" was used to test a null hypothesis of 1:1:1 for the gonotrophic states of nectar-feeding females. A chi-square contingency analysis was used to establish the relationship between parity and the type of feeding activity that the mosquitoes were engaged in (Steel and Torrie 1960). These tests were performed on the sums of unequal sample sizes and are thus subject to bias if the population base was changing during the sampling period. The degree of variability in the gonotrophic state and parity of individual samples is shown in Figs. 1, 2 and 3.

RESULTS

Totals of 492 females and 958 males were collected while probing or feeding on flowers. Most of the females were *Aedes vexans* Meigen (322) and *Culex restuans* Theobald (122). The others were *Ae. stimulans* (Walker) (1), *Ae. trivittatus* (Coquillett) (10), *Aedes* spp. (2), *Anopheles punctipennis* (Say) (13), *Culex* spp. (21) and *Psorophora ferox* (Humboldt) (1). Blood-seeking females comprised *Ae. vexans* (301), *Ae. trivittatus* (26), and *Ps. ferox* (1).

Of the *Ae. vexans* collected while nectar-feeding, most (78.6%) were empty, 3.7% were blooded and 17.7% were gravid (Fig. 1, Table 1). These proportions were all significantly different from one another ($P < 0.005$). The 12 blooded individuals comprised 7 freshly fed, 2 late fed and 3 subgravid. The parous/nulliparous (P/N) ratio of empty nectar-feeders was 0.53/1.00 (Table 2). Nearly all (97%) of the *Ae. vexans* collected while blood-seeking were empty; 2 were gravid and 8 (2.7%) were blooded (Table 1, Fig. 2). All blooded females were freshly fed, and 3 of these had ovarian follicles in stage III, indicating that they were taking a supplementary blood meal; the others presumably took blood shortly before being collected. The P/N ratio of empty blood seekers was 0.83/1.00 (Table 2). A comparison between parity ratios of blood-seeking and nectar-feeding mosquitoes showed that the type of feeding was related to parity ($P < 0.025$), though the difference was small.

Since the large samples from October 1 greatly influenced the results, parity ratios also were calculated without those samples. The P/N ratio of nectar feeders was 0.65/1.00; that of blood seekers was 1.93/1.00. A comparison of these values shows that the type of feeding was strongly related to parity ($P < 0.001$).

Among the nectar-feeding *Cx. restuans*, 47.5% were empty, 3.3% were blooded, and 49.2% were gravid (Fig. 3, Table 1). The proportion that was blooded was significantly different from the proportions empty and gravid ($P < 0.005$) and the proportions empty and gravid were not significantly different from each other ($P > 0.05$). The 4 blooded individuals comprised 2 freshly fed, 1 late fed, and 1 half-gravid. The P/N ratio of empty females was

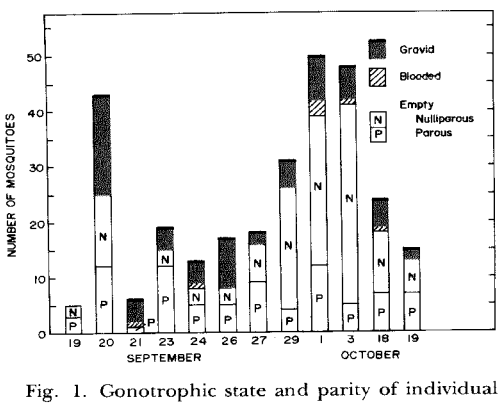

Fig. 1. Gonotrophic state and parity of individual collections of nectar-feeding *Aedes vexans*.

Table 1. Gonotrophic states of female mosquitoes collected while feeding on nectar or seeking blood.

Species	Behavior	Gonotrophic state			Total
		Empty	Blooded	Gravid	
Ae. vexans	Nectar feeding	253 (79%)	12 (4%)	57 (18%)	322
	Blood seeking	291 (97%)	8 (3%)	2 (1%)	301
Cx. restuans	Nectar feeding	58 (48%)	4 (3%)	60 (49%)	122

Table 2. Parity of empty female mosquitoes collected while feeding on nectar or seeking blood.

Species	Behavior	Parity			Ratio P/N	Total
		Nulliparous (N)	Parous (P)	Undetermined		
Ae. vexans	Nectar feeding	155 (61%)	82 (32%)	16 (6%)	0.53/1.00	253
	Blood seeking	151 (52%)	125 (43%)	15 (5%)	0.83/1.00	291
Cx. restuans	Nectar feeding	42 (72%)	12 (21%)	4 (7%)	0.29/1.00	58

0.29/1.00. Blood-seeking females for comparison were not obtained.

The 48 nectar-feeding females of miscellanous species and unidentified specimens comprised 28 empty, 2 blooded and 19 gravid specimens. Their respective numbers, according to genus, were as follows: *Aedes:* 10, 0, 3; *Culex:* 7, 2, 12; *Anopheles:* 10, 0, 3; *Psorophora:* 1, 0, 0. The P/N ratio of all empty specimens combined was 0.86/1.00. Of 7 *Ae. trivittatus* nectar feeders, 4 were parous (1.33/1.00), whereas of 25 empty *Ae. trivittatus* blood seekers, 21 were parous (5.25/1.00).

DISCUSSION

An underlying assumption of this study is that the samples of *Ae. vexans* and *Cx. restuans* females collected from flowers were representative of all nectar-feeding females of those species, at least in the study area in early autumn. This is a reasonable assumption, but difficult to assess. The sampling period was broad enough to cover periods both when the population of *Ae. vexans* was relatively old (high P/N ratio) and when there was a large influx of young females (low P/N ratio). Snakeroot and goldenrod, the only plants from which nectar-feeding mosquitoes were collected, were the most abundant plants flowering in the area at the time. Asters were also in bloom, but no mosquitoes were seen on them. The time of the collection period appeared to span the time of nectar-feeding activity. Our preliminary observations in Ohio confirmed the conclusion of Grimstad and DeFoliart (1974, 1975) that in Wisconsin, *Ae. vexans* and *Cx. restuans* (as well as many other temperate species) feed on nectar almost entirely in the 2 hr period after sunset, regardless of season or species of plant used as a nectar source. At our study site blood feeding activity of *Ae. vexans* was underway before sunset and diminished to a negligible amount by the time nectar feeding had ceased, in accord with the observations of Wright and Knight (1966). It appears that nectar-feeding may be one of sev-

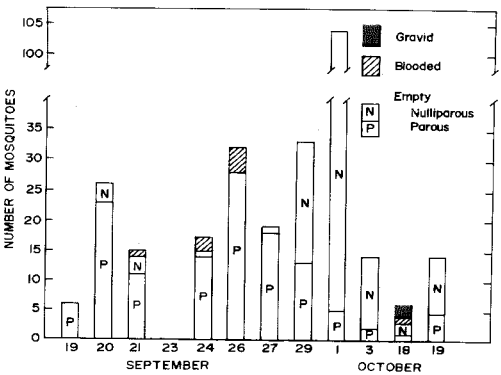

Fig. 2. Gonotrophic state and parity of individual collections of blood-seeking *Aedes vexans*.

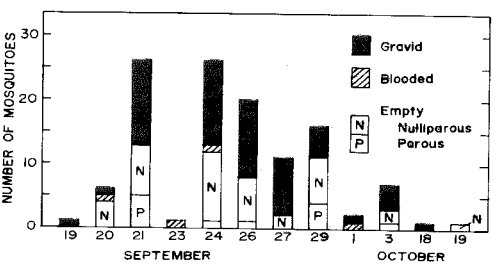

Fig. 3. Gonotrophic state and parity of individual collections of nectar-feeding *Culex restuans*.

eral behavioral options available to *Ae. vexans* during this activity period. Therefore, the presence of a vertebrate host (the collector) at the flowers may have influenced the composition of the *Ae. vexans* females (but not the ornithophilic *Cx. restuans*), by attracting those preferring a blood meal. However, like other workers (e.g., Magnarelli 1977), we have noticed that mosquitoes already probing or feeding on flowers seem not to be diverted to a host.

Mosquitoes that were digesting a blood meal were rarely found on flowers, regardless of species. This might be because there were few blooded mosquitoes in the population at large, or because they were less likely to take nectar when in that state. The latter is in accord with laboratory studies on *Ae. aegypti* showing that blood in the midgut inhibits responsiveness to extracts of milkweed flowers (Vargo and Foster 1982).

The proportions of gravid mosquitoes feeding on flowers were strikingly different between *Ae. vexans* and *Cx. restuans*, and also between the miscellaneous *Culex* spp. and the miscellaneous *Aedes, Psorophora,* and *Anopheles*. The predominance of empty females, which we observed in *Ae. vexans,* was likewise found in *An. implexus* (Theobald) by McCrae et al. (1976) in Uganda. Similarly, Magnarelli (1977, 1978) found that gravid resting *Aedes* spp. seldom contained fructose, a constituent of nectar, whereas empty females commonly contained it. By contrast, the nectar-feeding *Culex* mosquitoes we collected were as likely to be gravid as empty. This suggests that either the gravid state was not as likely to prevent *Culex* from nectar feeding, or there was a much higher proportion of gravid females in the *Culex* population. Frequent nectar feeding by *Cx. restuans* is a presumed characteristic of the hibernating generation in autumn (Wallis 1959), though it is doubtful that reproductively active mosquitoes also are affected in this way. It may be that there were fewer oviposition sites available to *Cx. restuans* in the collection area, so they were less likely than *Ae. vexans* to lay eggs before taking a nectar meal. This interpretation was used by de Meillon et al. (1967) to explain the difference in the proportion of gravid *Cx. quinquefasciatus* Say feeding on sugar cane trash during the dry season (11%), when breeding is favorable, and during the rainy season (64%), when it is not.

The presence of a substantial proportion of parous mosquitoes among the empty nectar feeders confirms the data of Magnarelli (1977, 1978, 1980), indicating that sugar feeding occurs not only soon after emergence and before the first blood meal, but also before subsequent ones. Since blood appears to be an inadequate source of energy for long-term survival of several mosquito species, and sugar feeding generally enhances egg production (Nayar and Sauerman 1971, 1975a, 1975b), the necessity of repeated feedings on nectar is evident. If nectar feeding occurs with the same frequency before and after the first blood meal, there should be the same amount of nectar feeding in nulliparous and parous mosquitoes. The data of Magnarelli (1978, 1980), based on fructose-positive rates of resting and blood-seeking females of several species, indicate that this is often the case. Our data on *Ae. vexans,* based on a comparison of the P/N ratios of nectar-feeding females and of blood-seeking females (representing the population at large), show a bias toward more nectar feeding among nulliparous individuals. This is especially obvious when the data of October 1 are removed. Apparently, a large emergence of adults occurred around this time. However, nulliparous females are probably underrepresented in the blood-seeking samples, because newly emerged mosquitoes commonly take sugar earlier than blood (W. A. Foster, unpublished observations). We tentatively conclude that *Ae. vexans* feeds on nectar one or more times before its first blood meal and continues to take nectar throughout adulthood.

ACKNOWLEDGMENTS

We are grateful to Dr. M. E. Clay, Dr. D. L. Denlinger and Maj. Harold J. Harlan for reviewing the manuscript and to Mr. Frederick S. Ruland for statistical advice and analysis.

References Cited

Brantjes, N. B. M. and J. A. A. M. Leemans. 1976. *Silene otites* (Caryophyllaceae) pollinated by nocturnal Lepidoptera and mosquitoes. Acta Bot. Neerl. 25:281–295.

Christophers, S. R. 1911. The development of the egg follicle in anophelines. Paludism 2:73–88.

de Meillon, B., A. Sebastian and Z. H. Khan. 1967. Cane-sugar feeding in *Culex pipiens fatigans*. Bull. W.H.O. 36:53–65.

Detinova, T. S. 1962. Age-grouping methods in Diptera of medical importance. W.H.O. Monogr. Ser. 47, 216 p.

Grimstad, P. R. and G. R. DeFoliart. 1974. Nectar sources of Wisconsin mosquitoes. J. Med. Entomol. 11:331–341.

Grimstad, P. R. and G. R. DeFoliart. 1975. Mosquito nectar feeding in Wisconsin in relation to twilight and microclimate. J. Med. Entomol. 11:691–698.

Hocking, B. 1953. The intrinsic range and speed of flight of insects. Trans. R. Entomol. Soc. London 104:223–345.

Jones, J. C. and B. V. Madhukar. 1976. Effects of sucrose on blood avidity in mosquitoes. J. Insect Physiol. 22:357–360.

McCrae, A. W. R., P. F. L. Boreham and Y. Ssenkubuge. 1976. The behavioural ecology of host selection in *Anopheles implexus* (Theobald) (Diptera, Culicidae). Bull. Entomol. Res. 66:587–631.

Magnarelli, L. A. 1977. Nectar feeding by *Aedes sollicitans* and its relation to gonotrophic activity. Environ. Entomol. 6:237–242.

Magnarelli, L. A. 1978. Nectar feeding by female mosquitoes and its relation to follicular development and parity. J. Med. Entomol. 14:527–530.

Magnarelli, L. A. 1980. Bionomics of *Psorophora ferox* (Diptera: Culicidae): seasonal occurrence and acquisition of sugar. J. Med. Entomol. 17:328–332.

Mer, G. G. 1936. Experimental study of the development of the ovary in *Anopheles elutus* (Dipt. Culic.). Bull. Entomol. Res. 27:351–359.

Nayar, J. K. and D. M. Sauerman, Jr. 1971. The effects of diet on life-span, fecundity and flight potential of *Aedes taeniorhynchus* adults. J. Med. Entomol. 8:506–513.

Nayar, J. K. and D. M. Sauerman, Jr. 1975a. The effects of nutrition on survival and fecundity in Florida mosquitoes. Part 1. Utilization of sugar for survival. J. Med. Entomol. 12:92–98.

Nayar, J. K. and D. M. Sauerman, Jr. 1975b. The effects of nutrition on survival and fecundity in Florida mosquitoes. Part 2. Utilization of a blood meal for survival. J. Med. Entomol. 12:99–103.

Nayar, J. K. and D. M. Sauerman, Jr. 1975c. The effects of nutrition on survival and fecundity in Florida mosquitoes. Part 3. Utilization of blood and sugar for fecundity. J. Med. Entomol. 12:220–225.

Nayar, J. K. and E. Van Handel. 1971. The fuel for sustained mosquito flight. J. Insect Physiol. 17:471–481.

Philip, C. B. 1943. Flowers as a suggested source of mosquitoes during encephalitis studies, and incidental mosquito records in the Dakotas in 1941. J. Parasitol. 29:328–329.

Shroyer, D. A. and D. P. Sanders. 1977. The influence of carbohydrate-feeding and insemination on oviposition of an Indiana strain of *Aedes vexans* (Diptera: Culicidae). J. Med. Entomol. 14:121–127.

Steel, R. G. D. and J. H. Torrie. 1980. Principles and procedures of statistics. A biometrical approach. McGraw-Hill, New York. 633 pp.

Vargo, A. M. and W. A. Foster. 1982. Responsiveness of female *Aedes aegypti* (Diptera: Culicidae) to flower extracts. J. Med. Entomol. 19:710–718.

Wallis, R. C. 1959. Diapause and fat body formation by *Culex restuans* Theobald. Proc. Entomol. Soc. Wash. 61:219–222.

World Health Organization. 1975. Manual on practical entomology in malaria. Part II. Methods and techniques. W.H.O. Offset Publ. No. 13, W.H.O., Geneva. 191 p.

Wright, R. E. and J. L. Knight. 1966. Effect of environmental factors on biting activity of *Aedes vexans* (Meigen) and *Aedes trivittatus* (Coquillett). Mosq. News 26:565–578.