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## THE SECOND BLOOD MEAL OF THE NORTHERN HOUSE MOSQUITO: BIRD VS. HUMAN BLOOD<sup>1, 2</sup>

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**ABSTRACT.** A comparison of host feeding behavior on chickens and humans was conducted with a Bladensburg, Maryland strain of *Culex pipiens* L. during 1977 and 1978. Three thousand female *Cx. pipiens* were tested for multiple feeding potential on chickens while 3200 *Cx. pipiens* were used for comparing multiple feeding preferences on chickens and hu-

mans. Data showed "opportunistic" feeding tendencies; 7.2% took the 2nd blood meal from a chicken; 4.0% from a man. It is suggested that this strain of *Cx. pipiens* has good vector potential for St. Louis encephalitis since, after taking bird blood, the second blood meal sometimes consisted of human blood.

### INTRODUCTION

An important factor governing the vector capability of a mosquito is its propensity toward taking a 2nd blood meal. It appears that little is known about the 2nd blood meal of mosquitoes in general and *Culex pipiens* L. in particular.

Among the reports on the feeding behavior of *Cx. pipiens*, the northern house mosquito, are those of Tate and Vincent (1936), Reeves and Hammon (1944),

Tempelis and Reeves (1964), Murphey et al. (1967), Tempelis et al. (1967), Means (1968), Kokernot et al. (1969), Hess and Hayes (1970), Ekis (1971), Spielman (1971), Tempelis (1975), and Magnarelli (1977). Most of these studies used precipitin tests to detect blood sources. All of them provided evidence that *Cx. pipiens* is primarily a bird feeder.

Edman and Downe (1964) found that *Cx. pipiens* "complex" females in Kansas fed on mammals more often than on birds. In their study 5 of 24 single feedings consisted of human blood with the remainder beef and hog. Tempelis and Reeves (1964) reported some feeding on man in Colorado and Illinois. In Delaware Murphey et al. (1967) found that *Cx. pipiens* fed on 7 species of small mammals. In the Mississippi River Basin Kokernot et al. (1969) found that 4 of 813 specimens tested had fed on man. Spielman (1971) sampled 194 specimens in Massachusetts and found 4 human feedings. Reeves and Hammon (1944) in Washington State

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found no human-fed *Cx. pipiens*, and negative results were reported by Tempelis et al. (1967) in Colorado. Means (1968) suggested that there were 2 different populations of *Cx. pipiens* on Long Island, New York: a farm population which was primarily ornithophilic and a woodland population which attacked man readily.

Precipitin testing yields very little information about sequential or multiple feeding. Edman and Downe (1964) pointed out that multiple feeding in precipitin test studies has for the most part been ignored. In their own studies they reported that the multiple feed rate of *Cx. pipiens* was 20% of all feeds recorded. Tempelis (1975) found multiple feeding to be uncommon. Spielman (1971) reported 2 double-fed *Cx. pipiens* in Massachusetts. One contained human and dog blood, and the other combined bird and mouse blood.

In 1975 there were 3 confirmed cases and 1 suspected case of St. Louis encephalitis (SLE) in the Bladensburg, Maryland area (Mallack et al. 1976). It was assumed that the northern house mosquito was the chief agent for transmission of the virus from birds to humans. (See Bailey et al. 1978.) Knowledge of the feeding habits of the suspected vector population has public health significance. The principal objective of the work reported here was to determine whether or not *Cx. pipiens* females which had taken bird blood would accept human blood for their second blood meal. We planned to obtain information about the relative acceptability of bird and human blood to mosquito females whose first blood meal had consisted of bird blood.

## MATERIALS AND METHODS

In September 1976 ca. 1000 *Cx. pipiens* larvae were brought from a truck rut in the Bladensburg area to the laboratory. The collection site in 1975 and 1976 was a focal point of SLE virus activity as indi-

cated by human cases, isolations from mosquitoes (Mallack et al. 1976) and sentinel chickens (Altman et al. 1977). In the insectary temperature and RH were kept at ca. 26°C and 70% respectively. Larvae were reared in enamel pans in distilled water with Gainesmeal<sup>®</sup> dog pellets. Pupae were placed in aluminum cages measuring ca. 61 x 61 x 61 cm. Adults were given 10% sugar water on cotton wads. White leghorn chickens provided blood. A chicken, secured with tape, was placed in the colony cage at intervals of 3 to 4 weeks, and large numbers of egg rafts were collected from the surface of distilled water in culture dishes. Viability of eggs averaged ca. 80%. The average cycle from egg to adult was ca. 17 days. The light/dark timing was as follows: 15 hrs daylight, ½ hr morning and evening twilight, and 8 hrs darkness.

A darkened laboratory area was used for the feeding experiments. The light/dark timing was the same as above, but the schedule was changed for convenience so that day and night were reversed. Temperature and RH in the experimental area were kept at ca. 25°C and 50% respectively. Group replicates of 200 females were allowed 48 hrs to adjust to the reversed day/night cycle. The 1st chicken blood meal, 1 hr duration, was offered at an average of 11 days post-emergence. Engorged mosquitoes were aspirated from each cage and placed in a plastic holding container, ca. 2 liter capacity, and given sugar water. These mosquitoes were held for ca. 2 weeks. Survivors were aspirated from holding containers to cages measuring 61 x 61 x 61 cm. and offered a 2nd blood meal for 1 hr.

A chicken was offered for the 2nd blood meal to 15 replicates. Sixteen replicates were used in the bird—human tests. For the 2nd blood meal offering, the investigator placed an arm in the cage for 1 hr. Mosquitoes that had not fed were aspirated into another cage and immediately were offered the chance to feed on a chicken for 1 hr.

Table 1. Acceptability of chicken and human blood to *Culex pipiens*, 1 hr. offering.

	First Experiment		Second Experiment		
	1st meal, chicken	2nd meal, chicken	1st meal, chicken	2nd meal, man	2nd meal, chicken
No. females	3000	949	3200	893	957
Avg. no./replicate	200	63	200	56	ca. 56
Avg. age in days	11	19	9	22	22
Range in age (days)	7-14	14-25	3-14	18-28	18-28
No. feeds	1508	68	992	36	7
% feeds	50.3	7.2	31.0	4.0	0.8
Deaths	559	N.A.	99	N.A.	N.A.

## RESULTS

Three thousand *Cx. pipiens* females in groups of 200 were offered chicken blood for 1 hr. Of this number, 1508 or 50.3% took a blood meal (Table 1). Subsequent offering of a 2nd chicken blood meal to 949 surviving females resulted in a feeding rate of 7.2%.

In the bird-to-man experiment 3200 females in groups of 200 were offered a chicken for 1 hr. Of this group 992 or 31.0% took chicken blood (Table 1); 893 survivors were offered human blood. Thirty-six or 4.0% took human blood. Mosquitoes which had rejected human blood after 1 hr were immediately offered chicken blood as a 2nd blood meal. Seven of 857 (0.81%) took chicken blood.

Incidentally we observed that most feeding on chickens occurred on the legs rather than on a greater area of exposed skin free of feathers. Mosquitoes offered a human arm tended to seek corners and crevices formed by the interface of skin and clothing or the cage.

## DISCUSSION

The precipitin tests reported by Edman and Downe (1964), Tempelis and Reeves (1964) and Spielman (1971) provide evidence that individual *Cx. pipiens* females do feed on both birds and mammals. However, the nature of the sequence, that is, the identification of the 1st and 2nd blood sources cannot be made by precipitin tests, and there is the suspicion that

multiple feeds represent atypical interrupted feeding. The host preference investigations of Means (1968) showed that *Cx. pipiens*, in seeking blood, discriminated among caged birds, reptiles, amphibians, and mammals; but only a single blood meal was studied.

Our results, namely a double feeding rate of 7.2% bird-to-bird and a bird-to-man rate of 4.0% for *Cx. pipiens* appear to be unprecedented. In fact very little published information about the 2nd blood meal of any species is available. In one of our tests the 0.8% bird-to-bird incidence suggests that those females definitely preferred bird blood inasmuch as they had refused mammal blood. On the other hand, in a fairly large proportion of the population opportunistic feeding tendencies were evident. It can be assumed, on the basis of percentages just given, that chicken blood for the 2nd blood meal was ca. twice as acceptable as human blood. We infer that in a suburban area where birds and men coexist the natural feeding behavior of the Bladensburg population of *Cx. pipiens* is such that this species may be considered a good vector of arboviruses from bird to bird and bird to man.

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