A THERMAL HEAT SUMMATION MODEL TO PREDICT THE DURATION OF THE GONOTROPHIC CYCLE OF *CULISETA MELANURA* IN NATURE

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ABSTRACT. This study determined the effect of temperature on the gonotrophic cycle of *Culiseta melanura* and developed a thermal heat summation model to calculate its duration under field conditions. A colony of *Cs. melanura* was used from New Jersey (F_{13}–F_{17} generation) and the length of the gonotrophic cycle was observed at 2, 10, 16, 22, 28, 32, and 34°C. None of the mosquitoes survived at 2°C or 34°C and none laid fertile eggs at 32°C. A linear regression analysis on the data showed that the thermal minimum for ovarian development was 6.4°C and 95.87 degree days were required above 6.4°C to complete one gonotrophic cycle. A thermal heat summation model is presented to allow calculation of the duration of the gonotrophic cycle under field conditions when average temperatures are known.

This study was conducted to determine the time required by *Culiseta melanura* (Coq.) to complete the first gonotrophic cycle under a range of temperatures. *Culiseta melanura* is the enzootic vector of eastern equine encephalomyelitis (EEE) virus in North America (Morris 1988, Scott and Weaver 1989, Crans et al. 1994). The time required to find a host, digest a fresh blood meal, and lay eggs is essential to understanding the full potential of this species as an amplifier of virus among bird populations in epizootic foci.

We used a New Jersey strain of *Cs. melanura* colonized by Mahmood and Crans (1994) in the F_{13}–F_{17} generations. Newly emerged adults were held in a 2 × 2 × 2-ft. cage in an insectary maintained at 22 ± 2°C with a 16:8-h (L:D) photoperiod. Because unmated females frequently take longer to digest a blood meal and develop eggs (Downe 1975), mating was ensured by providing a 1.5-h dusk and dawn crepuscular period. Adults were continuously provided with a 10% sucrose solution. To determine the optimum age for host acceptance, small groups of newly emerged females were offered a restrained northern bobwhite quail (*Colinus virginianus*) every night until bloodfeeding was evident. In these tests, newly emerged females generally took their first blood meal 7 days after emergence, which agrees closely with the observations of Morris (1984).

To determine the duration of the gonotrophic cycle over a range of temperatures, 6–9-day-old females were allowed to blood feed overnight on a restrained bobwhite quail. The following morning, groups of 20–45 fresh bloodfed females were isolated in 1-gallon cages constructed from ice cream cartons. The bloodfed females were provided with a 10% sucrose solution and the cages were transferred to an environmental chamber maintained at a 16:8-h (L:D) photoperiod and 80% RH. Mosquitoes were checked daily and females with stage IV eggs (Christophers 1911) were individually isolated in vials containing a small amount of deionized water for oviposition and a 10% sucrose solution. Vials were checked daily for egg rafts. The rafts were examined daily for 3 wk for evidence of hatching and were recorded as viable if they hatched under the temperature regime of the subset. The time required from engorgement to oviposition was determined at 2, 10, 16, 22, 28, 32, and 34°C. The duration of the gonotrophic cycle was calculated only from females that laid eggs that hatched.

Differences in the mean duration of the gonotrophic cycle due to temperature were tested using a one-way analysis of variance (ANOVA) and Duncans's new multiple range test ($P < 0.05$) (Sokal and Rohlf 1969).

A thermal heat summation model of the form $V = (t - t_0)/k$ (Mahmood and Reisen 1981) was calculated from the times required by *Cs. melanura* to complete one gonotrophic cycle over the range of temperatures used in these experiments. The thermal heat summation model was calculated using regression analysis between temperature (t) and rate of development (V). In this model, $V = 1/g_c$, where g_c = mean time in days from eggs in stage I (Christophers 1911) to oviposition. The regression of V on t must first be obtained as $V = a + bt$, where a is the intercept or rate of development when $t = 0$ and b is the slope of the regression line. In this model, k is used as a thermal constant or the number of degree days above t_0, the empirical thermal minimum below which ovarian development is arrested. The value k can be calculated as $1/b$ and t_0 can be obtained as $-(a/b)$.

Table 1 shows the effect of temperature on the duration of the gonotrophic cycle under controlled conditions. The amount of time required for complete blood meal digestion and oviposition decreased significantly with each increment in temperature from 10 to 28°C ($F = 648.17$; df = 3, 89; $P < 0.05$). Egg development was arrested at the 2°C temperature regime and only 5 of 20 females were alive after 72 h of exposure to this temperature. The females that survived developed normal

Table 1. Effect of temperature on the duration of the gonotrophic cycle of *Culiseta melanura*.

Temper- ature (°C)	Total females (isolated)	Total females (ovi- posited)	Gonotrophic cycle (days) (mean ± SD)[1]
2	20	0	ND[2]
10	20	3	23.2 ± 3.06a
16	26	25	11.5 ± 1.27b
22	45	41	5.7 ± 0.48c
28	33	24	4.5 ± 0.20d
32	25	4	ND
34	25	0	ND

[1] Means with different letters are significantly different by Duncan's new multiple range test ($P < 0.05$).

[2] ND = Length of gonotrophic cycle not determined because females either did not lay eggs or eggs were not viable.

egg rafts when the adults were transferred to 22°C. At 10°C, *Cs. melanura* took 16–20 days to develop their eggs to stage V but only 3 of 20 females in this group laid viable eggs. Blood meal digestion may have been hampered at 10°C, as some females retained small amounts of undigested blood after 20 days. At 16°C, 96% of the females survived and all laid fertile egg rafts within 10–12 days. At 22°C, 91% of the females survived to oviposit and egg development took only 5–6 days. At 28°C the gonotrophic cycle was completed in slightly over 4 days but 28% of the females died before they laid eggs. At 32°C only 16% of the females survived to lay eggs and none of the rafts hatched from females

held at this temperature. No bloodfed females survived to lay eggs at the 34°C temperature regime.

A linear regression analysis on the data showed that egg development in *Cs. melanura* is accelerated by an increase in temperature ($r^2 = 0.98$). The empirical thermal minimum (t_0) below which ovarian development is arrested in *Cs. melanura* was calculated as 6.4°C from the regression equation. The value of the thermal constant k or the number of degree days required above t_0 for completion of the gonotrophic cycle in these tests was 95.87 degree days.

Our calculations show that the thermal heat summation model for *Cs. melanura* is $g_c = 1/V$ where $V = (t - 6.4)/95.87$. This model can be used to calculate the duration of the first gonotrophic cycle as well as subsequent gonotrophic cycles in field-collected females.

Figure 1 is a graph generated from the thermal heat summation model showing the number of degree days above the thermal limit (t_0) generated by average temperatures from 7.5 to 28.5°C and corresponding durations for the gonotrophic cycle of *Cs. melanura* over those temperature ranges. The graph can be used to determine the duration of the gonotrophic cycle of *Cs. melanura* at any point in the breeding season where the average temperature ranges between 10 and 28°C. Because *Cs. melanura* females required at least 7 days for reproductive maturity at 22°C and are known to bloodfeed at this age, the first gonotrophic cycle would take 7 days in addition to the g_c value for the appropriate tem-
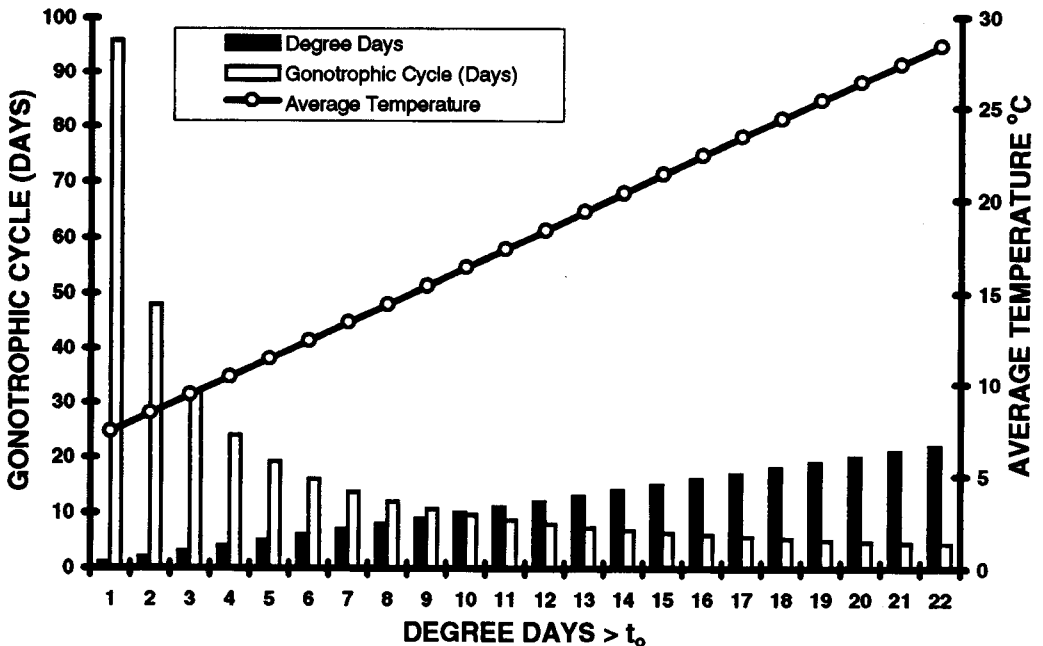

Fig. 1. Relationships among average temperature, number of degree days above the thermal minimum (t_0) required for egg development, and the duration of the gonotrophic cycle of *Culiseta melanura*.

perature. The maturation period should be excluded from calculations for subsequent cycles.

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