POPULATION SIZE, PARITY STRUCTURE, AND WING LENGTH OF *COQUILLETTIDIA PERTURBANS* IN AN OHIO FOCUS OF EASTERN EQUINE ENCEPHALITIS

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ABSTRACT. Adult female density, parity status, and wing length were determined weekly for a population of *Coquillettidia perturbans* in an area enzootic for eastern equine encephalitis virus in central Ohio. Samples were collected in CO_2-baited CDC miniature light traps from the first week in June through the 2nd week of September 1992. Population density indicated a single emergence peak during the 2nd week in July. However, parity rates showed 2 peaks, occurring in the first week of August (70.9% parous) and the 2nd week of September (55.3% parous), which suggested that there was a relatively small 2nd generation. Average wing length declined significantly over the season. The decline in size was negatively correlated with average air temperature occurring at least 6 wk before the time of emergence. Despite the seasonal decline in wing length, the low coefficient of variation for the average wing length (5.5) indicated relatively little variation in size. Comparison of parous and nulliparous female wing lengths each week suggested that there was no association between size and survival in this species.

INTRODUCTION

Inland foci of eastern equine encephalitis (EEE) virus transmission occur sporadically in the eastern half of the United States in association with swamp habitats harboring populations of *Culiseta melanura* (Coq.) (Morris 1988). Repeated isolation of EEE virus from *Coquillettidia perturbans* (Walker) has incriminated this species as a primary epizootic/epidemic vector in these areas (Howitt et al. 1949, Morris and Srihongse 1978, Francy 1982, Crans and Shulze 1986, Nasci et al. 1993). Isolation of EEE virus from *Cq. perturbans* has been associated with disease outbreaks in horses and humans in Michigan (Francy 1982) and Ohio (Nasci et al. 1993). During the 1991 EEE epizootic in Wayne and Holmes counties in east-central Ohio, the minimum infection rate of EEE in *Cq. perturbans* was calculated at 4.3/1,000 specimens tested. Except for observations of larval and adult density, little is known about the population biology of this species in midwestern EEE foci. The purpose of this study was to examine patterns in adult population density, parity structure, and body size in *Cq. perturbans* in the EEE focus in central Ohio.

MATERIALS AND METHODS

The study was conducted in the Killbuck Marsh Wildlife Area in Wayne and Holmes counties, Ohio, which was the center of the equine epizootic in 1991 (Nasci et al. 1993). Specimens were collected with dry ice-baited CDC miniature light traps from the first week of June through the 2nd week of September 1992. Traps were placed in the 12 locations used for collections during the 1991 epizootic. Once each week, 20–24 traps were run from approximately 2 h before sunset to 2 h after sunrise, except for the first and 2nd weeks in June and the 2nd week in August. During the first week of the June and the 2nd week of August, 24 traps were run each night for 6 consecutive nights to obtain large numbers of specimens for early- and late-season parity dissections and virus isolation attempts. No collections were made during the 2nd week of June. Daily minimum and maximum air temperatures were obtained from the Ohio Agricultural Research and Development Center (Ohio State University) in Wooster, Ohio, located 6 km north of the study site. The central tendency of weekly temperatures was calculated as the average of the daily (minimum + maximum)/2 for the week.

Specimens were frozen quickly in the field on dry ice. Mosquitoes were identified to species and sex on chill tables kept at 4°C. The ovaries were removed from at least 50 females from each trap-night (up to 5 *Cq. perturbans* females from each trap, each night). The ovarian tracheoles were observed to determine parity (Detinova 1962). In addition, the wing length of each of the dissected specimens was measured (distance from the axillary incision to the apical margin, excluding the fringe of scales) by using a binocular dissecting microscope equipped with an ocular micrometer. The remaining *Cq. perturbans* were pooled in groups of ≤100 and tested for the presence of arboviruses. Specimen

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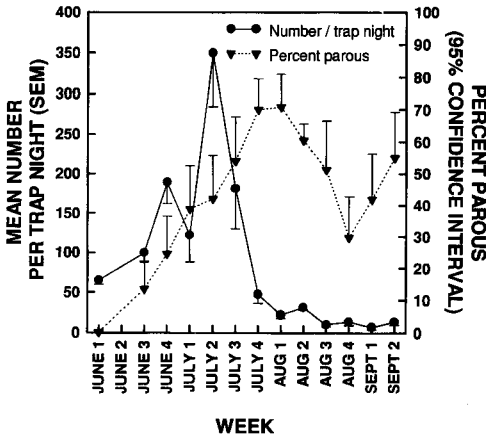
Fig. 1. Average adult *Coquillettidia perturbans* female density (number collected per trap-night) and percentage of females that were parous each week for specimens collected in the Killbuck Marsh Wildlife Area during 1992.
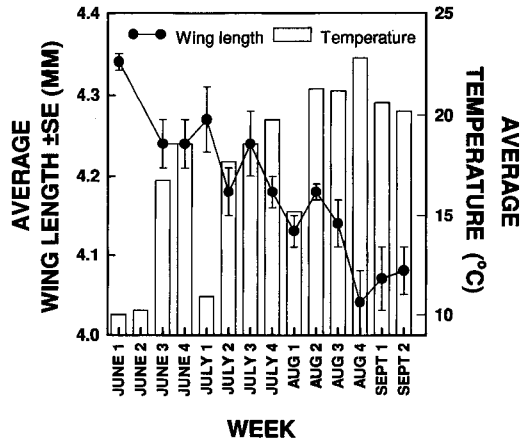

Fig. 2. Average wing length of *Coquillettidia perturbans* females collected in CDC light traps each week in the Killbuck Marsh Wildlife Area during 1992 and average temperature recorded 6 wk before collections were made.

preparation for virus identification was conducted as described in Nasci et al. (1993). Wing lengths of nulliparous and parous specimens were compared using a one-way analysis of variance (ANOVA); seasonal trends in wing length were analyzed with Pearson's product moment correlation. All statistical analyses were conducted as described in Sokal and Rohlf (1989).

RESULTS

Coquillettidia perturbans adult female density increased throughout the period from early June to mid-July, then declined rapidly and remained at relatively low levels for the remainder of the season (Fig. 1). On average, 145 females were collected per trap-night from the first week in June through the 3rd week in July. The peak emergence, 352 per trap-night, occurred during the 2nd week in July. During the latter part of the season (4th week of July through the 2nd week of September), we collected an average of 21 female *Cq. perturbans* per trap-night.

Altogether, 13,404 female *Cq. perturbans* in 448 pools were tested for the presence of viruses. Two pools collected on August 11, 1992 were positive for Jamestown Canyon virus, resulting in a minimum infection rate of 0.6/1,000 (2 positive/3,367 tested) during that week.

Of the 1,486 females dissected over the entire season, 65.2% were nulliparous, 33.0% were parous, 0.7% were gravid, and the parity status of 1.1% could not be determined. The seasonal pattern in parity structure is shown in Fig. 1. During the first week of June, 0.2% of the fe-

males were parous. The percentage that were parous gradually increased to a peak of 70.9% during the 2nd week of August, declined to 29.7% in the 4th week of August, then increased to 55.3% by the 2nd week of September.

The wing lengths of the 1,486 female *Cq. perturbans* that were dissected and measured ranged from 3.40 mm to 4.96 mm and averaged 4.23 ± 0.23 (SD) mm. The coefficient of variation (CV) of wing lengths in the population was 5.5, and the distribution was skewed to the left ($g_1 = -0.191$), indicating that the distribution of wing lengths was significantly different from a normal distribution (test for significance of a deviation from a parameter, $P < 0.01$).

The distribution of wing lengths over the season and the temperature 6 wk before the collection are shown in Fig. 2. There was a significant negative correlation between average wing length and week of the study ($r = -0.37$, $P < 0.001$), indicating that wing length decreased over the course of the season. Weekly average wing length and average temperature during the week of collection were not correlated (Table 1). However, there was a significant negative correlation with temperatures that preceded the collections by 6–12 wk (Table 1).

For the entire season, the average wing length of nulliparous females (4.25 ± 0.23 mm) was significantly longer than that of parous females (4.18 ± 0.22 mm) ($P < 0.001$, ANOVA). Average wing lengths of nulliparous and parous females by week are shown in Fig. 3. Nulliparous and parous female wing lengths were compared within each week to eliminate the impact of the seasonal reduction in body size. Nulliparous fe-

Table 1. Product-moment correlations between weekly average temperature and average wing lengths of female *Coquillettidia perturbans*.

	Time lag (weeks)								
	0	1	2	3	4	5	6	7	8
r^1	0.01	0.01	−0.53	−0.46	−0.49	−0.65	−0.76*	−0.59*	−0.83*

[1] Correlation coefficients followed by an asterisk (*) are significantly different from 0.

males had significantly longer wings during 1 week, the 2nd week in July ($P < 0.01$, ANOVA). Parous female wings were significantly longer during 3 weeks, the 2nd week in June, the 3rd week in July, and the first week in August ($P < 0.01$, ANOVA). There was no significant difference in wing length during the remaining 9 wk ($P > 0.05$, ANOVA).

DISCUSSION

The broad *Cq. perturbans* emergence peak observed during June and July in the Killbuck Marsh Wildlife Area is similar to patterns of emergence found in other populations of this species in the northern part of its range. Late spring to early summer emergences have been documented in Maryland (Saugstad et al. 1972), Massachusetts (Main et al. 1968), and Wisconsin (Siverly and DeFoliart 1968), as well as in areas to the north in Ontario (Helson et al. 1980, Allan et al. 1981) and the Nova Scotia–New Brunswick border (Lewis and Bennett 1980).
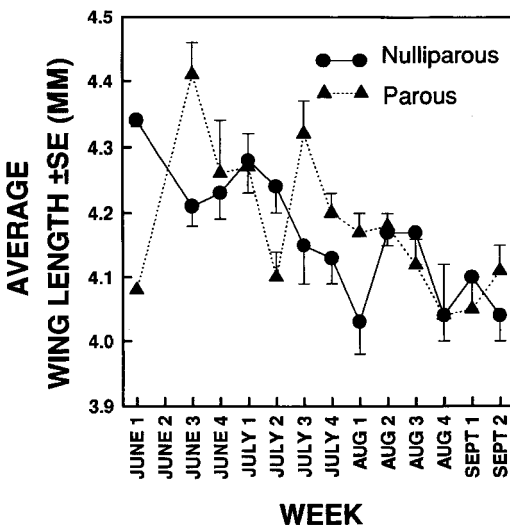

Fig. 3. Average wing length of parous and nulliparous *Coquillettidia perturbans* females collected each week during 1992 in the Killbuck Marsh Wildlife Area.

The population density of *Cq. perturbans* in the Killbuck Marsh Wildlife Area was considerably lower in 1992 than in 1991, when epizootic EEE transmission occurred in the area. During 1991, density exceeded 200 per trap-night in all collections from late May through mid-July (Nasci et al. 1993), whereas density during the 1992 spring emergence exceeded 200 per trap-night in only 1 week (2nd week in July). Thus, extremely high densities of this species may be required for epizootic transmission of EEE virus in inland foci (Crans and Schulze 1986). Because excessive early season rainfall is positively correlated with *Cq. perturbans* and *Cs. melanura* densities (Main et al. 1968), rainfall may be used as an early season indicator of the potential for epizootic EEE activity.

The overall parity rate of 33.0% was similar to the 26.9% found in a *Cq. perturbans* population in Wisconsin (Morris and DeFoliart 1971). However, the maximum parity rate of 70.9% was considerably higher than the 39% found in Wisconsin. This high parity rate in late season corresponds to the period of epizootic EEE virus transmission during the previous year (Nasci et al. 1993). The maximum parity rate in our study occurred 3 wk after the peak population density. Peak density and maximum parity were simultaneous in Wisconsin.

Although southern populations of *Cq. perturbans* appear to be either bivoltine, with peaks of emergence in June and September (Bidlingmayer 1968, Lounibos and Escher 1983), or trivoltine, with peaks in April–May, July, and September (Provost 1976, Slaff and Haefner 1985), previous studies suggest that this species is univoltine in the northern part of its range (Main et al. 1968, Siverly and DeFoliart 1968, Helson et al. 1980, Lewis and Bennett 1980, Olds et al. 1989). In the present study, the single, broad emergence peak, indicating a relatively synchronous emergence of adults at the beginning of the season, followed by a declining population density is consistent with a univoltine species. However, the decline in percentage parous from the 3rd week in August through the first week in September indicates a second adult emergence. This may represent a late season 2nd generation,

Table 1. Extended.

Time lag (weeks)			
9	10	11	12
−0.67*	−0.72*	−0.81*	−0.84*

which Allan et al. (1981) speculated could occur in years with warm summer and fall temperatures. Alternatively, this could represent a late emergence of individuals that overwintered in very early instars.

Seasonally declining body size, as indicated by decreasing wing length, is characteristic of patterns found in other species (Bock and Milby 1981, Landry et al. 1988, Lounibos et al. 1990, Willis and Nasci 1994) and appears to be related to temperature during larval development. Significant negative correlation of wing length occurs with temperatures 1 wk before trapping in *Culex tarsalis* Coq. (Bock and Milby 1981), 3 wk before trapping in *Mansonia dyari* Belkin, Heinemann and Page (Lounibos et al. 1990), and 1 month before trapping in *Aedes albopictus* (Skuse) (Willis and Nasci 1994). In the present study, the 6-wk lag between trapping and a significant negative correlation between temperature and wing length probably results from the very long larval development time of *Cq. perturbans* and the time required for water to warm in spring.

The 5.5 CV of average wing length indicated little variation in body size in this population of *Cq. perturbans*. This, and the significant negative skewness of the wing length distribution, places *Cq. perturbans* in the moderate size-variability group of Fish (1985) along with other mosquito species inhabiting relatively permanent larval habitats like swamps and lake margins, where predation is the major factor controlling population size.

Over the entire season, wing length and parity rate were negatively correlated, suggesting that smaller *Cq. perturbans* had higher survival rates. However, examination of the differences in size and parity each week throughout the season indicated generally no difference in wing length between parous and nulliparous females. Therefore, we concluded that wing length and survival in *Cq. perturbans* were not associated, probably because there was so little variation in wing length or body size in this species. This is likely the case in other species with low variation in size. The difference in wing length and parity detected in seasonal data was probably due to many nulliparous females emerging in spring, when average wing length was large.

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