# DIEL PATTERNS OF OVIPOSITION AND INFLUENCE OF AGITATED WATER SURFACE IN *CHIRONOMUS ANONYMUS* (DIPTERA: CHIRONOMIDAE)

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ABSTRACT. A series of experiments was carried out in a 0.5-ha lake at Portola Country Club, Palm Desert, CA, to study the oviposition habits of *Chironomus anonymus*. Egg masses were collected by floatable polypropylene ropes attached to the 1.5-m-long stakes placed in the lake. In the diel experiment, ropes were changed every 2 h to determine the preferred oviposit on periods of *C. anonymus*. The peak of oviposition was between 1900 and 2100 h. For the interruption of oviposition experiment, ropes were placed in the still and agitated portions of the lake for 24 h. A significantly higher number of egg masses, according to applied *t*-test, was collected from the ropes placed in the still portion of the lake.

## **INTRODUCTION**

Chironomus anonymus Williston, when emerging in large numbers from lakes and ponds in country clubs, resorts, or golf courses, poses a nuisance problem to residents and visitors. Currently, 2 preparations are registered for midge larval control, temephos (organophosphate [OP]) and diflubenzuron (insect growth regulator [IGR]) (Mulla 1974, Mulla et al. 1976). The microbial control agent Bacillus thuringiensis var. israelensis is effective against some groups of chironomid larvae, but is not yet registered for midge control (Mulla et al. 1990, Rodcharoen et al. 1991). Although these preparations have good levels of activity and low toxicity to nontarget organisms, they have certain use limitations. They are active against a small number of midge species and have high cost. Therefore, other strategies need to be incorporated in an effective integrated control for nuisance midges. One possibility is to interrupt the oviposition. Prior to implementing this strategy, it is necessary to determine the diel oviposition rhythms of the problem species.

Existing information about chironomid eggs and oviposition primarily deals with the ovipositon behavior of certain species in the laboratory (Chaudhuri and Nandi 1983, Ali et al. 1987, Xue et al. 1994), egg mass morphology (Ashe and Murray 1983), or furnishing a key to egg masses (Morrow et al. 1968). Experiments in the laboratory indicate a midge preference for a stable substrate bordering the water during oviposition (Ali et al. 1987). Observations in the field suggest the same preference of midges for a stable substrate during oviposition (Davies 1976).

The most common techniques for collecting

chironomid egg masses include various types of mesh cloth placed at the bottom of the water body (Morrow et al. 1968, Frommer and Rauch 1971), or a long string anchored on both ends and threaded through a number of floatable corks with attached filter paper strips (Credland 1973).

In our study we used polypropylene ropes attached to stakes to recover information about the oviposition habits of *C. anonymus*, and the interruption of oviposition.

## MATERIALS AND METHODS

The 7 lakes at the Portola Country Club had concrete edges, bottoms sealed with plastic liners, and were supplied with tertiary-effluent water. A detailed description of the lakes in Portola can be found in Lothrop et al. (1993). Studies were carried out in the lake at hole 10 of the golf course. The surface area of the lake was *ca*. 0.5 ha, with a maximum depth of 1.5 m. Two aeration fountains were placed in one end of the lake. For collecting egg masses, we used floatable 0.75-cm-diam polypropylene ropes. Each rope was tied with twine to a 1.5-m wooden stake and placed in the lake. The ropes floated at or just below the water surface.

The influence of color and length of ropes on the oviposition intensity of *C. anonymus* was determined by using 10 ropes of different color and length in 2 separate experiments. Two ropes of each length (10, 15, 20, 25, and 30 cm), one yellow and one bicolored red and white, were used. Two groups of rope lengths were formed to analyze the effect of length. The first group included 10-, 15-, and 20-cm ropes and 25- and 30-cm ropes. The ropes of each group were placed in still and agitated portions of the lake. Data were analyzed by *t*-test (Sokal and Rohlf 1969).

The diel oviposition rhythms were studied on 3 separate dates. On September 28, 1994, the

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Rope colors			No. of egg masses per rope								Total	
Yellow	386	476	1,112	1,437	1,187	865	272	4	12	35	5,786	
Red/white	411	1,314	989	269	530	518	77	26	406	807	5,347	

Table 1. Comparison of rope colors in Chironomus anonymus oviposition preference.<sup>1</sup>

 $t_{0.05(18)} = 1.73; t_{obs} = 1.30.$ 

oviposition experiment was accomplished only during the scotophase, from 1700 to 0700 h, because of increasing wind in the rest of the 24-h period. On October 12, 1994, egg masses were collected only during the photophase, from 0700 to 1700 h, because of increasing wind during the rest of the 24-h period. The continuous 24-h diel oviposition experiment was conducted on August 2–3, 1995. On each date, 5 yellow, 30-cmlong ropes were placed in the still portion of the lake, and changed every 2 h.

To study the effect of water turbulence on interruption of oviposition, water around the aeration fountains was used as the agitated portion of the lake, whereas the still area of the lake served as the control. Twenty-four-hour experiments were conducted on September 22–23 and 27–28, 1994, and on July 12–13, 1995. Ten yellow ropes, each 30 cm long, were used on each date. Five ropes were placed in the agitated portion and 5 in the still portion of the lake. The significance of agitated water as a method of interrupting oviposition was estimated by comparing the numbers of egg masses collected in the still and agitated portions of the lake by *t*-test.

The ropes with collected egg masses were stored in zip-lock bags with sufficient water for eggs to float, and transported to the laboratory. Egg masses were placed in a white enamel tray and counted. Some egg masses were placed in rearing containers and raised to the adult stage to confirm the species identity, and some were stored in 70% alcohol. The only species found at the time of experiments was *C. anonymus*.

Before each oviposition experiment, 5 submerged traps were placed in the lake to ascertain if the emerging adult population was sufficient to pursue the experiment. The technique for adult submerged trapping is described in Mulla (1974). Adults were stored in 70% alcohol and later identified after mounting in Euparal medium, following the procedure in Wiederholm (1989).

The larval density was estimated by taking 5 benthic mud samples each time before the oviposition experiment. The technique for taking mud samples and processing midge larvae is described in Lothrop and Mulla (1995).

# **RESULTS AND DISCUSSION**

An initial experiment was conducted to ascertain if rope length or color had affect on the rate of *C. anonymus* oviposition. According to the *t*-test analysis (df = 18), there was no significant difference between the yellow and bicolored red and white ropes ( $t_{0.05(18)} = 1.73$ ,  $t_{obs} = 1.30$ ) (Table 1). There was no difference between the first (10-, 15-, and 20-cm) and second (25- and 30-cm) group of rope lengths (df = 3) (Table 2). The ropes of each length group were placed in still and agitated portions of the lake ( $t_{obs} = 0.24$  for still and  $t_{obs} = 0.94$  for agitated,  $T_{0.05(3)} = 2.35$ ).

During the scotophase diel experiment (1700– 0700 h), sunset was at 1835 h and sunrise at 0639 h. Peak oviposition occurred between 1900 and 2100 h, with 2,206 egg masses collected. A smaller peak was obtained between 0500 and 0700 h, when 320 egg masses were collected (Fig. 1). Submerged trapping yielded 507 adults in 5 traps, and 928 larvae were counted in 5 mud samples the day before.

During the photophase diel experiment (0700–1700 h), sunset was at 1816 h and sunrise at 0649 h. Maximum oviposition occurred between 0700 and 0900 h, with 34 egg masses collected (Fig. 1). Submerged trapping collected 124 adults in 5 traps, and 2,480 larvae were counted in 5 mud samples the day before.

During the 24-h diel experiment, peak ovi-

Table 2. Effect of rope length on Chironomus anonymus oviposition preference.

Water type		1st roj	pe group		2n			
	10 cm	15 cm	20 cm	Total	25 cm	30 cm	Total	t <sub>obs</sub>
Still	804	406	989	2,199	1,112	865	1,977	0.24
Agitated	4	10	35	49	77	411	488	0.94

 $t_{0.05(3)} = 2.35.$ 

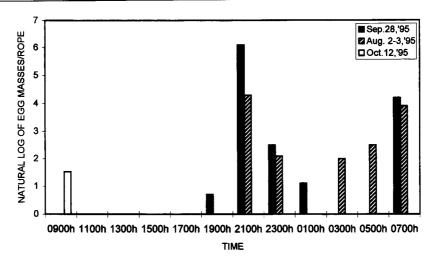


Fig 1. Diel oviposition activity of Chironomus anonymus.

position occurred between 1900 and 2100 h, with 77 egg masses collected (sunset was at 1947 h and sunrise was at 0557 h). A slightly smaller peak occurred in the morning, between 0500 and 0700 h, with 61 egg masses collected (Fig. 1). Submerged traps collected 158 adults in 5 traps, and 1,668 larvae were counted in 5 mud samples the day before the experiment.

Data on interruption of oviposition indicated a significant difference in the number of egg masses laid on the ropes placed in still and agitated sections of the lake (Figs. 2, 3, and 4). An applied *t*-test (df = 8) showed a significant difference in all 3 experiments ( $t_{obs(1)} = 2.14$ ,  $t_{obs(2)}$ = 2.03,  $t_{obs(3)} = 1.39$ , and  $t_{0.01} = 1.57$ ,  $t_{0.05} =$ 1.86). On September 22–23, 1994, about 19 times more egg masses were laid on the ropes placed in still water, whereas on September 27–28, 1994, the difference was about 3 times (Figs. 2 and 3). During the July 12–13, 1995, experiment there were no egg masses collected from the ropes placed in the agitated portion of the lake (Fig. 4).

Our results indicated that an agitated surface significantly impeded oviposition by *C. anonymus.* Most likely, water was washing over the ropes creating vulnerable surface for the act of oviposition, resulting in fewer observed egg masses. Davies (1976) noted that moderate winds can affect chironomid oviposition site selection, either by altering the midges' flight or by making otherwise suitable oviposition sites undesirable. Ripples created by aeration fountains, which are similar to those generated by

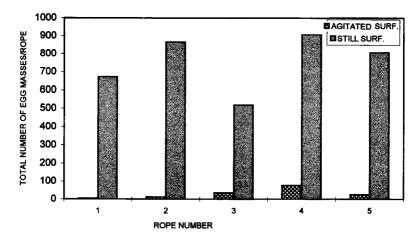


Fig. 2. Total number of *Chironomus anonymus* egg masses laid per 5 ropes in the still and agitated portion of the lake on September 22-23, 1994.

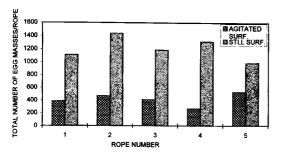


Fig. 3. Total number of *Chironomus anonymus* egg masses laid per 5 ropes in the still and agitated portion of the lake on September 27–28, 1994.

moderate winds, evidently influenced ovipositing midges in our study.

One way to create ripples on the water surface would be to place sprinklers around the lake edge. These sprinklers could be set to activate during the peak oviposition time of problem species. Another solution would be to increase the number of aeration fountains in the lakes. This nonchemical control measure targets the midge population at the egg level, reserving insect growth regulators and other chemicals for emergency use. However, before an integrated control plan incorporates interruption of oviposition as a standard method for midge control, it is essential to obtain more information on oviposition rhythms for the problematic species.

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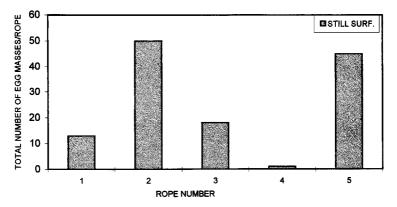


Fig. 4. Total number of *Chironomus anonymus* egg masses laid per 5 ropes in the still and agitated portion of the lake on July 12–13, 1995.

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