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PRE-FABRICATED DINING SHELTERS AS OUTDOOR INSECTARIES, AN ASSESSMENT USING *ENALLAGMA EBRIUM* (HAGEN) (ZYGOPTERA: COENAGRIONIDAE)

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Problems with housing adult damselflies include unnatural responses to being caged and prohibitive costs of outdoor cages. Here, a relatively inexpensive method, using pre-fabricated tents as outdoor insectaries, is presented. These tents do not adversely affect flying, foraging, mate searching, and mating of damselflies. Survival rates of pre-reproductive adults were also high (100% survival of teneral females for the entire pre-reproductive period in 1 of 2 trials). Outdoor insectaries also have been used successfully for housing other spp., incl. *E. boreale* Sel. and *Nehalennia irene* (Hag.).

INTRODUCTION

Adult and larval odonates are often used as model organisms for testing ideas about evolution and ecology (e.g., FINCKE, 1988; JOHNSON, 1991). Many tests of theory, however, require experimental approaches. Whereas larval odonates are amenable to experiments, adult odonates are less useful in this regard. Larval odonates are amenable to experiments because they can be housed indoors in relatively simple containers and provided with various foods which are easily available, such as microcrustaceans like *Daphnia* or polychaete (enchytraeid) worms (BAKER et al., 1992). Many field trials of housing larval odonates also have been conducted (review by JOHNSON, 1991). Adult odonates are not housed so easily. Some studies have been successful in housing damselflies indoors in relatively simple containers for breeding experiments (JOHNSON, 1964; CORDERO, 1990), whereas in other studies, wings of zygopterans were cut, which precluded animals

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from flying indoors, and animals had to be fed by hand (HINNEKINT, 1987).

Outdoor insectaries could prove useful for large-scale experiments on adult damselflies (or perhaps dragonflies). Outdoor insectaries could allow researchers to vary densities or sex ratios of animals, while providing a semi-natural setting in which duration and intensity of certain behaviours are compared between experimental treatments. Importantly, there are only a few references available on the use of outdoor insectaries for housing zygopterans (FINCKE, 1987; FORBES, 1991). The problem immediately apparent in using insectaries is expense especially if a large space is required for vagile animals, or if a large number of replicates is required for statistical rigor. In addition, there is always the problem of unnatural behaviours as a response to caging.

This paper presents a relatively inexpensive method of housing flying insects through the use of pre-fabricated tents, or 'dining shelters', as outdoor insectaries. These dining shelters are meant to cover outdoor tables to allow people to remain outdoors without being bothered by mosquitoes or other insects. We examined survivorship and behaviour of *Enallagma ebrium* (Hag.) with regards to efficacy of using such tents as outdoor insectaries. The experiments described herein were part of a larger study on the effects of parasites on survivorship and reproductive success of *E. ebrium* adults (FORBES, 1991b).

METHODS

PROJECT 1. Pre-reproductive survival of captive *E. ebrium* males and females

Two 'dining shelters', marketed by the Canadian Tire Corporation (model no. 75-5118-2), were erected (in early July 1989) in an old field near Chaffeys Locks, Ontario Canada (44°34' N; 79°15'W). This field was on land owned by Queens University; *E. ebrium* adults were observed to forage and roost in this field. These tents were made of polyethylene mesh, supported by aluminum poles, and shaped like truncated pyramids. The tents measured 3.65 x 3.65 m at the base, 3.00 x 2.75 m at the top, and were roughly 2.0 m high (Fig. 1). Tents cost ca $50 CDN each at the time of this study.

Fig. 1. A pre-fabricated tent, as an outdoor insectary.

One of us (MF) removed all large arachnids and insects including other damselflies (Coenagrionidae), dragonflies (Libellulidae), spiders (Araneidae), harvestmen (Phalangiidae), ambush bugs (Phymatidae), and robber flies (Asilidae) from tents before introducing newly-emerged male and female *E. ebrium* on 12 and 14 July, 1989 (Trials 1 and 2, respectively). These large insects were likely competitors or predators of *E. ebrium*. In total, 16 male and 20 female teneral *E. ebrium* were released into each tent.

Each adult *E. ebrium* held in captivity was marked uniquely following HINNEKINT (1974) on the first day after capture. An assistant made 10-20 sweeps with an aerial insect net (diameter 0.5 m) in vegetation outside the tent every 2 days. Small insects including leaf hoppers (Cicadellidae), spittlebugs (Cercopidae) and ants (Formicidae) (>200 individuals for 10-20 sweeps) were removed from nets and released into tents. After 1 week had passed, we entered inside tents from 11:00-13:00 hours every 2-3 days. During visits, dead or moribund animals were collected. Moribund animals were those animals that were unable to fly or crawl away after being lightly touched. We removed these animals before ants disposed of dead or dying damselflies. Two males for each trial could not be accounted for. These males may have escaped from the tent during our entry or may have died and been carried off by ants. No data were obtained for these males.

As a secondary objective, we examined whether parasitism by *Arrenurus magnicaudatus* (Marshall) mites influenced survivorship of male or female *E. ebrium*. FORBES & BAKER (1991) demonstrated that these mites are associated with reduced survivorship of mature male *E. ebrium*, but no work has examined survivorship of captive damselflies over their pre-reproductive period in relation to ectoparasitism. We examined whether mean numbers of *Arrenurus* mites differed between those animals which died before versus after mid-date (the date by which 50% of the experimental animals had died). Each sex-by-trial combination was considered separately.

PROJECT 2. Mating behaviour of captive *E. ebrium*

As a second project, two tents were erected at the border of Upper Dowsley: a pond near the Queen's Biological Station described by FORBES (1991a). Two coenagrionid species were commonly encountered at this pond: *E. boreale* (Sel.) and *E. ebrium*. These two species had flight seasons that overlapped. *E. boreale* started emerging in late May and flew until early July; at that time, *E. ebrium* started emerging and was commonly seen mating at the pond by middle of July, 1989 (FORBES, 1991b).

We examined responses of mature males and females released into tents. We wanted to determine whether mature males that were mate-searching at sites could be captured and released into tents at the same site, without greatly affecting their natural behaviours. For females, we captured tandem pairs at the edge of the pond and gently removed the male. We then stored females in holding cages, before releasing them into tents. Holding cages measured 42 x 30 x 30 cm. These cages were provided with nylon meshing on the top and three sides, whereas the square bottom was plywood (1.25 cm thick). A plexiglass sliding door formed one side of the cage.

As before, tents were cleared of all potential competitors or predators, before adding *E. ebrium* adults. However, we did not release prey animals into these tents. Males and females were housed in tents for only a few hours on any day, and then released. To determine how many males to put into tents, we staked four 3 x 3 m plots at the edge of the pond. We recorded how many males were present in those plots from 13:00-14:00 h on 16, 18 and 20 July. On average, 11 ± 1.76 males were present in those 3 x 3 m plots. Thus, we added 15 males to tents to simulate natural densities as the floor space of these tents was slightly larger than the plots (i.e. 3.65 x 3.65 m). Males were added to tents on each of 5 occasions (18, 20, 22, 25, and 27 July).

Males were allowed to become accustomed to tents for 0.5 h before releasing 2-6 females (by gently sliding the plexiglass door on the holding cage). We recorded interactions between males and females; we were mainly interested in whether copulations were observed and whether they were of similar duration to those seen in natural settings (FORBES & TEATHER, 1994). As before, individuals were marked uniquely, so that they could be identified should they become lost in the melee of mate searching males and/or tandem pairs within the tent.

RESULTS

PRE-REPRODUCTIVE SURVIVAL OF CAPTIVE *E. EBRIUM* MALES AND FEMALES

Mortality rates during the pre-reproductive period were similar for males and females in trial 1. However, in trial 2, females suffered no mortality (Fig. 2). Importantly, females were found in copula with surviving males at the end of both trials.

To examine whether mean ($\log_{10}$ (x+1)) number of mites differed between animals dying before versus after mid--dates, we used t-tests preceded by F-tests for homogeneity of variances (for females in trial 1: $F_{9,9}=1.05$; for males in trial 1 $F_{6,6}=1.55$; for males in trial 2: $F_{6,6}=1.57$; all $p > 0.05$). Our results show that pre-reproductive mortality was unrelated to the degree of parasitism by mites for both males and females (Tab. I).

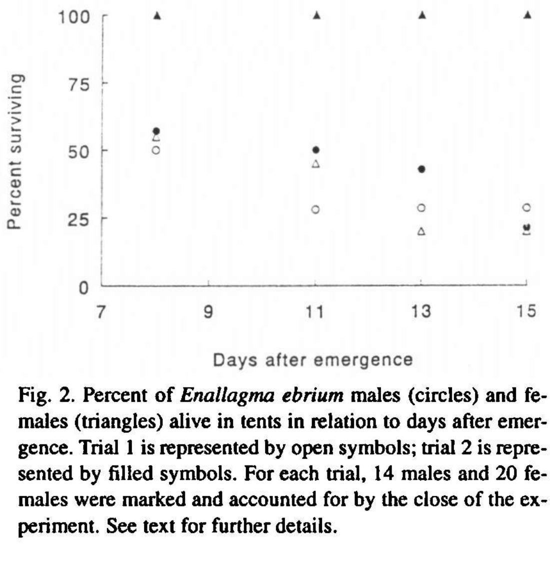

Fig. 2. Percent of *Enallagma ebrium* males (circles) and females (triangles) alive in tents in relation to days after emergence. Trial 1 is represented by open symbols; trial 2 is represented by filled symbols. For each trial, 14 males and 20 females were marked and accounted for by the close of the experiment. See text for further details.

Table I

Mean numbers of mites on *E. ebrium* males and females dying before, versus living after, the date by which half of the experimental animals had died. Females from trial 2 are omitted because none were observed to die after 15 days. T-tests were based on transformed data ($\log_{10}$(x+1)). – See text for details of experimental procedure

Trial	Sex	N	Status	Mean	T-Value	P
1	m	7	alive	11.64		
					0.238	ns
		7	dead	10.74		
	f	10	alive	11.88		
					0.699	ns
		10	dead	9.71		
2	m	7	alive	8.77		
					0.279	ns
		7	dead	8.31		

MATING BEHAVIOUR OF CAPTIVE *E. EBRIUM*

Over all experimental releases, 17 females were captured by males and mated with males in tents (3 of 4 females released on 18 July, 2 of 2 females released on 20 July, 1 of 3 females released on 22 July, 5 of 6 females released on 25 July and 6 of 6 females released on 27 July). Thus, 33-100% of females released in tents mated within the first hour of being released. For 6 pairs, we were able to record complete copulation durations, that were preceded by sperm translocations by males (see ROBERTSON & TENNESSEN, 1984, for a description of this behaviour). Mean copulation duration was 32.7 ± 9.53 min.

DISCUSSION

E. ebrium of both sexes flew normally in tents (they did not 'blunder' into walls) and both sexes foraged on leafhoppers and ants while in tents (M. Forbes, pers. obs.). Our results suggest that retention in tents with abundant food (and no predators) was not particularly stressful because some females collected after the pre-reproductive period in the first project were mated to surviving males and contained developed eggs, and also because all females survived the pre-reproductive period for the second trial of the first project. In addition, we found that naturally--occurring levels of parasitism by larval water mites did not affect pre-reproductive survival of male and female damselflies when housed in semi-natural field enclosures with abundant food.

Adult males and females collected at the edge of the pond and released into tents would often mate quickly. All of the females released into tents were captured by males, indicating that males were able to mate-search effectively in tents. However, some females simply refused to mate (see FORBES, 1991, for a description of female behaviours that indicate 'unwillingness' to mate). That some females did not remate may be due to the experimental protocol of catching tandem or copulating pairs, removing males, housing females in containers, and later releasing those females. Some females may respond less well to such handling. Alternatively, there may be natural variation in a female's willingness to remate that is not indicative of 'abnormal' behaviours resulting from experimental and housing protocols. Importantly, copulations observed in this study were nearly equal to those reported for natural matings of *E. ebrium* (32.7 versus 29.4 min; this study versus FORBES & TEATHER, 1994, respectively).

We believe that tents described in this paper can be used as outdoor insectaries, but that researchers will have to take care designing experimental protocols. We also housed *E. boreale* Sel. and *Nehalennia irene* (Hag.), each in a single tent over 10 and 5 days, respectively. We observed matings between conspecifics at the end of both time periods. However, in a related experiment in which *E. ebrium* adults were housed in tents some 25 m away from the edge of a site (see FORBES, 1991a, for a description of this site), males were not observed to mate search, but rather flew at the wall of the tent in the direction of the site. Perhaps these males were simply removed too far from the water's edge. Thus, care should be taken in determining placement of tents, e.g., animals that were mate-searching may not be removed too far from the edge of ponds and expected to continue mate-searching behaviours.

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