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SMALL OUTDOOR INSECTARIES AS A TOOL FOR LIFETIME STUDIES ON DAMSELFLIES

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Abstract. Damselflies are suitable subjects for examination of a variety of biological questions, but most research has been carried out in the field. Several questions are hard to test because of the uncontrolled conditions inherent in field studies. This can be circumvented by studying populations in semi-natural outdoor insectaries. We assessed the suitability of such insectaries by comparing the survival and adult behaviour of *Ischnura elegans* (Vander Linden) in insectaries and in the field. Our results showed that damselflies behaved differently under experimental conditions. Nevertheless, outdoor insectaries can be regarded as a valuable tool to elucidate questions concerning life history traits since they offer the possibility to eleminate predation, emi- and immigration and hidden life.

Key words : Odonata, damselflies, insectaries, experimental setup.

INTRODUCTION

In the last two decades, it has become clear that damselflies are suitable model organisms to study a diversity of behavioural and ecological topics (*e.g.* FINCKE *et al.*, 1997). One of their advantages over vertebrates is their variety in reproductive strategies and the possibility they provide for the collection of lifetime data over a short timespan. Housing larval damselflies has been succesfully conducted by several researchers (see JOHNSON, 1991). In contrast most research on imagines has been performed in the field.

Several problems and limitations apply to conventional field studies. First, in the field up to half of a study population may disappear after marking and releasing (*e.g.* FINCKE, 1988 ; CORDERO, 1997). As a result, conclusions may not be based on a random sample of the population. A second problem is the occurrence of unknown successful dispersal to other ponds (MICHIELS & DHONDT, 1991a). Third, examining several populations simultaneously is difficult because of habitat complexity ; and problems (*e.g.* due to weather conditions) may arise when life history is studied in different periods or years (*e.g.* BANKS & THOMPSON, 1985). Fourth, observed frequencies of copulation and oviposition in the field are often an underestimation of the reality (VAN NOORDWIJK, 1978 ; CORDERO *et al.*, 1997). Finally, it is extremely difficult to control and vary the variables sex ratio, age distribution and density (but see SIVA-JOTHY, 1995) which are important for testing hypotheses (*e.g.*

MICHIELS & DHONDT, 1991a; FINCKE, 1994). Some of these problems have been avoided by observing populations in large outdoor cages (MICHIELS & DHONDT, 1991a; DUNHAM, 1994). However, testing more than two populations at the same time remains difficult. Here, we present an inexpensive method for housing damselflies in small outdoor insectaries, enabling the simultaneous study of several populations with chosen population ecological parameters.

A major possible criticism of this type of experiment is the possibility that unnatural behaviour may occur. Therefore we examined the differences between small insectaries and the field. We observed *Ischnura elegans* (Vander Linden) in insectaries and, simultaneously in the field, and compared our findings with literature data. The insectary experiments described herein are part of a broader study on sexual strategies in that species.

MATERIAL AND METHODS

Four dining shelters (Partytent, marketed by Bricomarkt B.V.B.A.; size: 3x3x2.5 m), were covered with bee-netting (mesh size: 2x8 mm, marketed by B.V.B.A. Ranschaert) (Fig. 1). In contrast with former studies (FINCKE, 1987; FORBES & LEUNG, 1995) the netting allowed passage of small insects and avoids the need to supply food. To allow enough light, we replaced triangular parts of the plastic roof with the bee-netting. Insectaries were secured against heavy wind by stretching tent-ropes and by digging in the undersides of the bee-netting. The total price of an insectary was approximately 2000 BEF (65 USD) at the time of the study.

Fig. 1. – Insectary.

Each insectary contained two small pools (plastic shells, Boubsy N.V.) with a perimeter of approximately four meter each, to serve as oviposition sites and supply of water on hot days. Around the pools we planted *Juncus effusus* as roosting sides (GEIJSKES & VAN TOL, 1983) The remaining vegetation consisted mostly of *Arrhenaterum elatius, Artemia* *vulgaris, Cirsium arvense, Dipsaccus fullonum, Plantago major* and *Urtica dioica* with heights between 20 and 80 cm.

The insectaries were placed in a field called « De Biotuin », property of the University of Antwerp. Experiments were performed during the summer of 1998. To avoid predation, the insectaries were cleared of spider webs and frogs. The animals used for the experiments were collected approximately 10 km away from our study site (« De Walenhoek », Niel). Captured animals were transported in cages (size : 30x30x30 cm). All animals were marked with an individual number in black ink on the left forewing (Staedtler Pancolor, 0.3 mm pen). Observations were made with binoculars from outside the insectaries (Opticron, 10 x 42).

In a first experiment we examined whether the food availability in the insectaries was sufficient to exlude mortality due to starvation and cannibalism (ROLFF & KRÖGER, 1997). We released 63 freshly emerged tenerals (34 males and 29 females) to one insectary and counted the number of survivors after maturation (five days). Survival of adults was analysed on a new group of animals in the second experiment. The combination of both experiments should produce an idea of the adequacy of the food available during the study.

In the second experiment we examined survival and adult behaviour including daily activity pattern and timing, duration and frequency of copulation and oviposition of adult males and females in four insectaries. We performed field studies on *I. elegans* during the summers of 1996 and 1997 (unpublished results), and compared both findings. We daily recorded interactions (tandem formation, copulation, oviposition) (from 8h00 until 18h00). Individuals in one insectary were observed during 15 min, whereafter we focused on the next insectary. Observations on insectaries were made in a daily random sequence. On three sunny days we observed damselfly activity between 7h00 and 8h00 at dawn. Hours are given in Mid European Summer Time.

Because of small sample sizes, we compared the daily activiy pattern between insectaries and the field by grouping frequencies of copulations and ovipositions per two hours. The analysis was performed using a Fisher Exact test. We analysed the effect of insectary and sex on the survival of adult *I. elegans* with a Cox proportional Hazard model (KLEIN & MOESCHBUERGER, 1997) using proc PHREG in SAS 6.12. Ties were handled with the EXACT procedure. Means are reported with one standard deviation.

RESULTS

Survival

In the first experiment we found that mortality of tenerals was low, and similar for males and females (respectively three and four damselflies died ; Fisher Exact test, df = 1, p = 0.69). We have no comparative field data. Survival of adult animals did not differ between insectaries and sexes (log likelihood test, (χ_2= 3.24, df = 4, p = 0.52 ; Fig. 2). Average adult lifespan was 17.67 ± 7.45 days. In the field study we observed an average lifespan of only 3.2 ± 0.5 days (unpublished results), without a difference in survival between males and females. We did not observe cannibalism in insectaries or field.

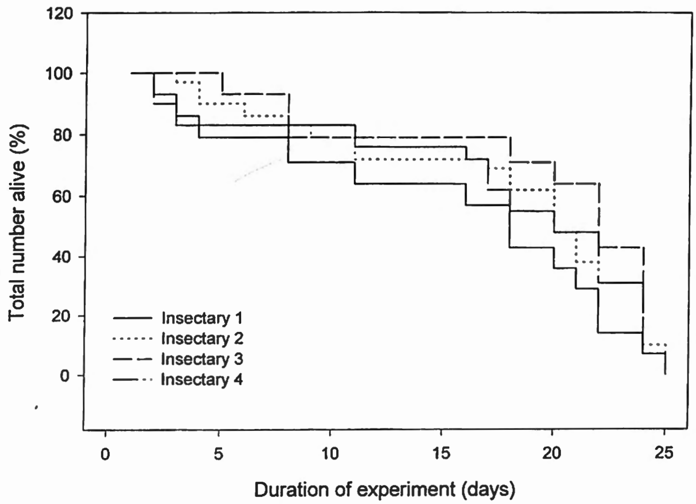

Fig. 2. – Survivorship curves of adult *Ischnura elegans* in outdoor insectaries.

Adult behaviour

At night animals roosted on the vegetation of the insectary. During the day, sunshine resulted in males making long flights, inspecting vegetation looking for females. Unwilling females prevented males from mating by spreading their wings and curling their abdomen. At the end of a copulation, pairs perched in tandem for periods ranging from 1 min up to an hour. Long post-copula tandems were mostly observed close to oviposition sites. After post-copula tandem, animals separated and sometimes continued to perch alone at the same spot. Females oviposited without male mate guarding. They were aggressive towards intruding males or females, attacking, and sometimes even clashing with them, thereby temporarily reserving an oviposition site for their exclusive use. Field observations were similar, except that at night, damselflies were found roosting only on emergent aquatic or littoral vegetation.

Daily activity pattern

On sunny mornings, the first individuals began to flutter and to fly to the tops of the vegetation at 7h15 while the insectaries were shadowed and temperatures just above 15° C. Males grasped perching females and formed tandem linkages when sunshine entered the insectary. These tandems usually lasted a few minutes on a sunny morning but up to two hours when it was cloudy and/or rainy. The first copulations were observed at 8h05 and became numerous by 10h00. Most copulations were initiated before 11h00, except on cloudy days. When weather was bad, most activities were retarded by one to two hours except oviposition which, in contrast, occurred up to two hours earlier. Only in extremely bad conditions (harsh rain and strong wind) was no activity observed. Most oviposition took place from 16h00 onwards. Exceptionally, oviposition was observed earlier in the afternoon and only twice before 9h00. Under field conditions we observed a similar daily distribution of ovipositions (Fisher Exact test, df = 4, p = 1.0) (fig. 3). We found, however, a shift in the daily distribution for copulations (Fisher Exact test, df = 4, p = 0.034) whereby copulations in insectaries were initiated and terminated earlier (Fig. 3).

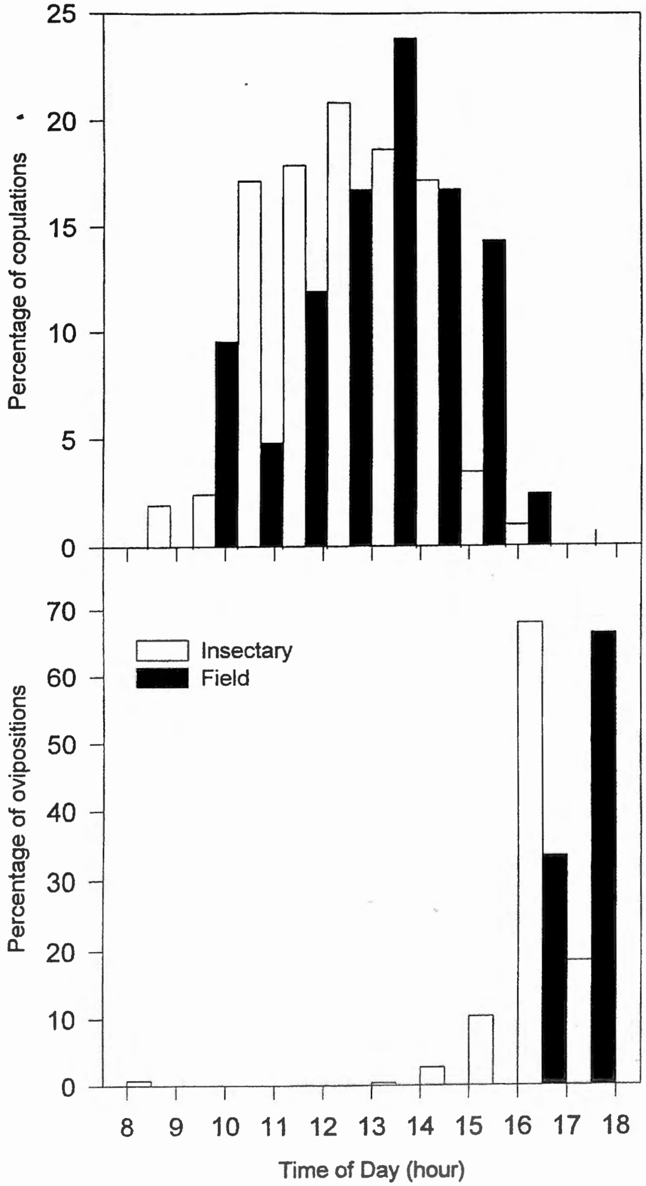

Fig. 3. – Daily distribution of copulations (upper panel) and ovipositions (lower panel) in adult *I. elegans* in insectaries and field.

Duration and frequencies of copulation and oviposition

Observed lifetime copulation frequency in the insectaries ranged from zero to twenty in males, and from zero to thirteen in females. Mean copulation time was 292 ± 19 min (N = 11). The maximum number of ovipositions observed per female was fifteen, while mean oviposition time was 38 ± 24 min (N = 17). Numbers of copulations and ovipositions were much lower in the field during summer of 1996 and 1997. Here, maximum number of matings was two for both males and females. In total, the field studies took more than a month and resulted in the observation of only three ovipositing females.

DISCUSSION

Both teneral and adult *I. elegans* foraged successfully on available prey. Mortality rates for tenerals were low and animals had enough energy to mate, while females were seen ovipositing regularly. Ovipositing in particular is very energy demanding due to the continuing development of eggs (*e.g.* ANHOLT *et al.*, 1991). Moreover, the survival of adult damselflies is generally higher in insectaries (LORD, 1961; VAN NOORDWIJK, 1978; HINNEKINT, 1987; CORDERO *et al.*, 1997). Therefore, our results suggest that stress due to food limitation was improbable in tenerals and adults. The use of bee-netting offers the advantage that we did not have to add insects (see FINCKE, 1987; LANGENBACH, 1993; FORBES & LEUNG, 1995) or supply rotting fruit to the insectaries (see FINCKE, 1987). In contrast with most studies on teneral survival (*e.g.* THOMPSON, 1989), predation pressure could be kept close to zero. Only occasionally a damselfly was trapped in a spiderweb.

The daily activity pattern of *I. elegans* was similar in insectaries and in the field (MILLER, 1987; HILFERT & RÜPPELL, 1997; see results and Fig. 3). The differences observed in the distribution of copulations can probably be attributed to varying weather conditions between the periods of examination. Copulation duration observed in this study was comparable with those reported for matings of *I. elegans* in the field (KRIEGER & KRIEGER-LOIBL, 1958: up to 340 min; MILLER, 1987: 324 ± 90 min). On the other hand, copulation frequencies in the insectaries were clearly higher than in the field. Low mating frequencies in the field are a general phenomenon in damselflies. CORDERO *et al.* (1997) found a maximum of three copulations per lifetime for a male and seven for a female, and PARR & PALMER (1971) found even lower numbers. The same can be said for oviposition frequency. The most likely explanation for this difference between insectary and field is the difference in detection probability due to the complexity of natural habitats compared to our insectaries, where we restricted the vegetation to a minimum. Indeed many copulations and ovipositions, even in insectaries, were hardly visible. Another aspect of lower observed copulation frequencies in the field is that many copulations occur away from the water (VAN NOORDWIJK, 1978; CORDERO *et al.*, 1997). Finally, the higher observed copulation frequencies may also be a consequence of the higher encounter rates between the sexes in encaged conditions (*e.g.* MICHIELS & DHONDT, 1991b).

The higher observed mating and oviposition frequency offers a great advantage. It increases the possibility to find differences between individuals or groups of individuals.

Moreover, the observation of all copulations and ovipositions is required if one wants to study a fitness component like lifetime reproductive success.

An additional advantage of working with insectaries is the possibility to eliminate or introduce mortality due to predation. In the field, predation often strongly reduces the variance in phenotypic characters, which makes it difficult to detect any sexual selection upon them (WADE & KALISZ, 1989). While one solution to circumvent this is to artificially increase the variance in the population for a character upon which both survival and sexual selection might act (*e.g.* Anholt 1991), another is to retain the variance in the population and differ the predation pressure. The latter can be achieved in insectaries where the researcher can examine a range of predator frequencies in different insectaries. Moreover, in the described small insectaries, several populations can be monitored simultaneously while controlling factors such as sex ratio and population density to test specific hypotheses regarding their effects on fitness. Therefore, we conclude that outdoor insectaries are a powerful tool to study questions concerning life time characteristics of damselflies.

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