

## Hand feeding: a method to increase the survival rate of *Orthetrum coerulescens* (Odonata: Libellulidae) in outdoor enclosures

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Food intake rate and diet composition have a high impact on all organisms and affect individual fitness, fecundity and mortality. Specimens in enclosures have to be fed in an adequate way and with minimum stress for the specimens. Adult dragonflies are flying hunters. In enclosures, they are usually fed by adding different kinds of adult dipterans. In this study, I additionally fed specimens of *Orthetrum coerulescens* by hand. Each specimen received one house fly or up to six fruit flies per day. This was less than an adult dragonfly would normally consume; however, this additional hand feeding was enough to significantly increase the survival rate of individuals, especially within the first 12 days of adult life. The maximum life span (subadult and adult) observed was about 60 days, for both hand-fed adults and those that were not hand fed.

**Keywords:** Odonata; dragonfly; Libellulidae; *Orthetrum coerulescens*; survival rate; maturation period; hand feeding; life span

### Introduction

Food intake rate and diet composition have a high impact on individual fitness, fecundity ([Laurila, Kujasalo, & Ranta, 1997](#); [Peckarsky, Cowan, Penton, & Anderson, 1993](#); [Scrimgeour & Culp, 1994](#)) and lifespan ([Lee et al., 2008](#)). Odonata are opportunistic foragers in their larval and adult stages. The usual diet of adults typically consists of small insects, predominantly Diptera ([Beatty, 1951](#); [Corbet, 1999](#)). For some studies, it is necessary to keep adult odonates in enclosures. For example, some damselfly species have been reared under laboratory conditions ([Cordero, 1994](#); [van Gossum, Sánchez, & Cordero Rivera, 2003](#); [van Gossum, Stoks, & de Bruyn, 1999, 2005](#)) and dragonfly species in semi-natural outdoor enclosures ([Hassan, 1977](#); [Michiels & Dhondt, 1989, 1991](#)). [Van Gossum et al. \(1999, 2005\)](#) used gauze with a large mesh size for the outdoor enclosures, allowing small insects to enter the enclosure naturally. However, this also increases the risk of spider predation ([Hassan, 1977](#)). Alternatively, fine mesh around the enclosure reduces the predation risk. Different kinds of dipterans and small butterflies can be added to provide a varied diet, as would be found in nature. [Van Gossum et al. \(2003\)](#) observed that the dipterans crowded in the corners of the enclosures and were out of reach for the odonates. This lack of food can cause nutrient deficiency and shorten life span ([van Gossum et al. 2003](#)). Hand feeding can perhaps compensate for this nutrient deficiency and help to reduce the high mortality

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during the maturation period when odonates are kept in enclosures. To my knowledge the effects of hand feeding have not yet been documented in odonates. In this study I tested whether hand feeding is feasible and increases survival of immature dragonflies in enclosures. Specimens of *Orthetrum coerulescens* were reared in outdoor enclosures with two different diet scenarios: (a) a large quantity of dipterans in the enclosure; and (b) additional hand feeding of one house fly or six fruit flies per specimen per day. Additionally, I investigated whether females laid fertilized eggs under these semi-natural conditions.

## Material and methods

I constructed two outdoor enclosures ( $4\text{ m} \times 4\text{ m} \times 2\text{ m}$ ) using two timber scaffolds covered with fine white gauze (1 mm mesh size). The bottom of the enclosures was covered with 1 mm thick foil, which was covered with a layer of sand. Each enclosure contained four large tub plants for perches. The enclosures stood next to each other and were partly covered with cotton fabric to shelter them from direct rain and sun. On days with temperatures above  $28^\circ\text{C}$  and no precipitation, I moistened the gauze of the enclosures with water from a hose once per day.

At the end of April 2009, I caught 190 larvae of *O. coerulescens* in the Canal de Vergiere, a small canal situated in the stony steppe of the Crau in Southern France, 25 km south-east of the city of Arles ( $43^\circ 34' \text{ N}, 4^\circ 50' \text{ E}$ ). I transferred the penultimate instar larvae into several plastic basins ( $0.4 \times 0.4 \times 0.2 \text{ m}$ ; 10 larvae per basin) filled with pond water and wooden stakes for emergence substrates, and placed these in outdoor enclosures. The water was changed weekly and larvae were fed twice a week with chironomid larvae *ad libitum* plus a random mixture of small macroinvertebrates.

Every day the enclosures were checked for emerged and dead adult specimens. When handling the adult specimens it was important to have no cosmetics like hand cream, body milk or sun lotion on the hands. Several hours after emergence individuals were numbered consecutively on the forewing using a black permanent marker (Staedtler lumocolor, Staedtler, Nuremberg, Germany). Specimens marked with an even number were hand fed every day with one house fly or, if this was not taken by the dragonfly, with six fruit flies (the weight of six fruit flies is equal to that of one house fly; hereafter I will use fruit flies as standard unit). Both treatments (hand fed and not hand fed) and both sexes were distributed equally over the two enclosures.

In both enclosures, hundreds of flies (fruit flies *Drosophila hydei* (Sturtevant 1921), house flies *Musca domestica* Linnaeus 1758, and flesh flies *Calliphora* sp.) were present and were offered as prey for the adult odonates. Oat porridge with organic waste material was provided as a food source and as egg laying and larval habitat for the dipterans. I had cultures both of flying and of flightless dipterans. The latter were used for hand feeding as they were easy to catch. In a preliminary experiment I also tried to hand feed the dragonflies with flesh flies; however this was not possible. I started the hand feeding one day after emergence. I caught the specimens of *O. coerulescens* with one hand and held their wings during feeding. I offered live dipterans to the dragonflies by presenting with forceps the dipterans head directly in front of the dragonfly's mandibles (Figure 1). Those specimens which did not accept any kind of prey offered one day, had to wait for the next feeding procedure on the following day. To analyse whether the survival rate differs significantly between the two treatments (hand fed and not hand fed) I used the Kaplan Meier test (Kaplan & Meier, 1958) (SPSS 20.0, IBM, New York).

Two weeks after emergence, when the specimens started to get their mature coloration, I tried to obtain egg clutches daily. Oviposition was initiated by dipping a female's abdomen into a jar filled with water (c.f. Boehms, 1971; Schenk, Suhling, & Martens, 2004). One day after oviposition I was able to distinguish between developing eggs and those that were not developing (see also Schenk & Söndgerath, 2005).



Figure 1. Hand fed female of *Orthetrum coerulescens*.

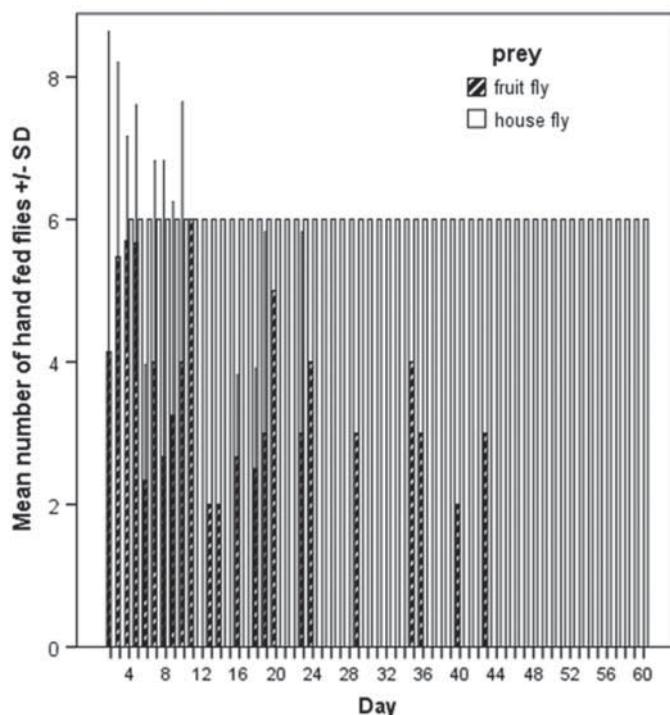


Figure 2. Mean number of flies (split between house and fruit flies), with standard deviation (SD), hand fed during the life span of the specimens of *Orthetrum coerulescens*. One house fly weighs as much as six fruit flies (see "Methods"); in this figure, therefore, one house fly was equated to six fruit flies.

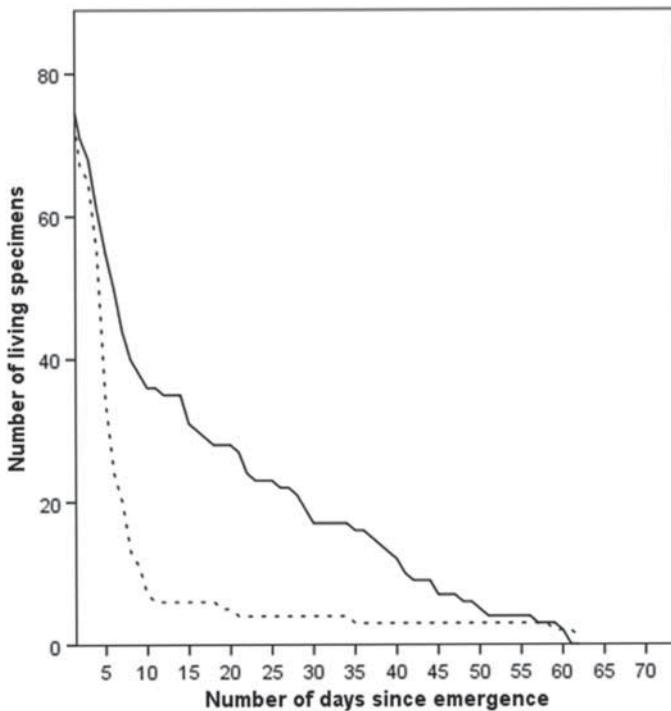


Figure 3. Survival rate of specimens of *Orthetrum coerulescens* fed by hand (continuous line) and those not fed by hand (dotted line).

## Results

In the outdoor enclosures, 81% of the specimens emerged successfully. The total number of subadult and adult specimens within the two enclosures changed constantly over the study time. Individuals emerged over two and a half months (23 May to 11 August 2009). A mean number of 11.21 specimens (subadult and adult)  $\pm 8.86$  SD per day occupied each enclosure during the study time. The sex ratio at emergence was female biased: 73 males to 81 females.

Dragonflies were observed feeding on all three dipteran species. I observed that until an age of 3–4 days after emergence the specimens fed mostly on small fruit flies. From the fourth to fifth days on they preferred to eat house flies. Regularly I observed mature adults hunting flesh flies. Similar to the scenarios described in van Gossum et al. (2003), the dipterans often crowded in the corner of the enclosures. This effect was reduced by disturbing the flies several times per day. Some of the semi-aquatic macroinvertebrates used as prey in the larval basins also hatched in the course of the experiment (Chironomidae, Coenagrionidae, Culicidae, Baetidae, Ephemeroptera, Lestidae, Tabanidae). I observed specimens of *O. coerulescens* taking them as additional food. This is an easy way to provide a more diversified diet.

Adults readily accepted hand held prey (Figures 1, 2). In only 9.7% of all hand feeding trials did the dragonflies reject fruit and house flies. Individuals took 5.3 fruit flies  $\pm 1.9$  SD as prey per feeding session. In the first three days of life the specimens did not eat the house flies offered. From the fourth day on house flies were accepted as food (Figure 1). The dragonflies' ability to fly was affected neither by the marking of the wings after emergence nor by the constant handling and feeding procedure.

The specimens of the two enclosures did not differ significantly (Mann–Whitney test) in their survival rate or life span. Therefore, I combined the data of both enclosures. Survival rate of

the dragonflies differed considerably between treatments: hand fed specimens had a significantly higher survival rates than the ones not hand fed (Kaplan Meier, Breslow = 0.53, df = 1,  $p$  = 0.022) (Figure 3). The maximum life span of around 60 days was equal in the two groups (Figure 3).

Hand feeding increased survival of the dragonflies, but this was a time-consuming activity. It took around an hour to feed 20 dragonflies with six fruit flies each. Feeding house flies accelerated the procedure; 20 dragonflies were fed in about 10 minutes in total. Hand feeding of other libellulid species (*Orthetrum cancellatum*, *Sympetrum striolatum* and *S. vulgatum*) with freshly caught specimens was successful as well.

During the study time I observed several copulations between adult *O. coerulescens*. After a mean of 23.46 days  $\pm$ 4.08 SD the females laid eggs for the first time. Although sometimes the last observed copulation happened hours or even days earlier, it was possible to get eggs from the females during the manually generated oviposition in the enclosure. The eggs of both hand fed and not hand fed *O. coerulescens* were fertile.

## Discussion

In general, all three dipteran species (fruit flies, house flies, and flesh flies) were accepted as hunting prey by *O. coerulescens*. For hand feeding only fruit flies and house flies were accepted. During hunting and hand feeding the choice of the food depended on the age of the dragonflies. Freshly emerged specimens preferred fruit flies. Medium-aged specimens preferred house flies. Older specimens preferred house flies and flesh flies for hunting and house flies for hand feeding. As the number of flies offered per day was much lower than dragonflies would normally eat, they only can be seen as supplementary food. Nevertheless, the hand feeding significantly reduced the mortality rate within the first 12 days of the maturation period. Increasing the number of flies offered per day might reduce the mortality rate even more. As hand feeding also worked with freshly caught specimens of *Orthetrum cancellatum*, *Sympetrum striolatum* and *S. vulgatum*, I assume this method can be used in general for libellulids.

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