TIME OF COLOUR CHANGE IN FEMALE *ISCHNURA PUMILIO* (CHARPENTIER) (ZYGOPTERA: COENAGRIONIDAE)

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Freshly emerged females were marked individually and put into an outdoor cage. By following the individual change of weight and width of abdomen it could be shown that the time of colour change is connected with maturation. The tested females were divided into 2 groups with different food supplies. The transformation of colour took less time in well-fed individuals than in less well-fed ones. In both groups the time for colour transformation was negatively correlated with the date of emergence and the sum of the daily maximum temperatures during the first six days of life.

INTRODUCTION

Ischnura pumilio lives in temporary or newly created waters from which it usually disappears after a few years (FOX, 1989; JURZITZA, 1970; RUDOLPH, 1979). This might be one of the reasons why the biology of this species is relatively unknown, although it is found almost everywhere in central, southern and eastern Europe, as well as in some localities in the British Isles, North Africa and Siberia (AGUILAR et al., 1985; ASKEW, 1988).

Female polymorphism is very common in the genus *Ischnura* and has been well studied in some species. It can take the form of true dimorphism, as in *I. gemina*, although the andromorphic female is rare in this species (GARRISON & HAFERNIK, 1981), or there might be just a colour difference due to age as in *I. hastata*, *I. posita* or *I. prognata* (DUNKLE, 1990), but in most cases it is both. For example, five colour variations are described for the female of *I. elegans*, two of which are teneral stages of the other three (KILLINGTON, 1924). Colour change due to age, as well as to true dimorphism has been described also in *I. verticalis*. (CALVERT, 1915), although FINCKE (1987) found only one

andromorphic female of this species. The same has been found for *I. pumilio* (BILEK, 1952; ZIMMERMANN, 1973). I found only three andromorphic females of *I. pumilio* during my study.

For a long time the orange female of *I. pumilio* (form *aurantiaca*) has been supposed to be a teneral stage (BILEK, 1952; CHAM, 1990; JURZITZA, 1970; KYLE, 1961; LONGFIELD, 1937; WELLINGHORST & MEYER, 1979; ZIM-MERMANN, 1973). BILEK (1952) was the first to observe colour transformation in captured females. CHAM (1990) was able to recapture green females which he had marked while they were orange. There is disagreement on the question of whether orange females are able to copulate or not. KYLE (1961) supposed that orange females were not able to copulate, but contrary to this JURZITZA (1970) and REINHARDT (1990) observed orange females in a copulation wheel and in tandem, respectively. The females of *I. aurora* are known to copulate shortly after emergence, and after copulation they emigrate to found new colonies (ROWE, 1978). In this paper I describe the influence of different factors on the maturation time of female *I. pumilio* with special reference to colour transformation.

MATERIAL AND METHODS

Observations were performed at the Domaine du Ligagneau, from June to August 1992. The area lies within the Camargue in southern France and has been recently converted into a nature reserve. The old irrigation system is now used to study the influence of irrigation and grazing on the vegetation of the Camargue. Thus a multifarious habitat has been created. *Ischnura elegans* is the most common zygopteran in this area, followed by the far less common *I. pumilio*.

Over a period of 25 days freshly emerged females (n=123) were collected, marked individually and kept in outdoor cages. A cage consisted of four wooden pegs rammed into the ground and a net $(2.05 \times 1.6 \times 1.7 \text{ m})$ extended over them. I used a green mosquito net of polyethylene with a mesh size of about 3 mm. The net was fixed at the ground by stones. A water-filled tub (about 40 l) was placed in the ground and planted with rushes (*Juncus maritimus*). The cages were sheltered from the wind. Individuals of *I. elegans* and *pumilio* were common in the surrounding area. At the beginning of the experiment the damselflies were not fed, but they were able to eat small insects entering the cage. On the 8th, 10th and 12th day and from then on daily I caught small flies (Ephydridae) at a nearby pool and put them into the cage to supplement their food supply. From the 23rd day on it was impossible to catch any more flies because the pool had dried out. This caused that one damselfly group (a) (n=92) was less-well fed than the other (b) (n=31). It is not possible to give more precise information about the food supply of group (a) because of its heterogeneity, some insects were additionally fed and others not. Neither can I say how much prey entered the cage by itself. Females in group (b) were fed daily, and had a surplus of food.

To protect the freshly emerged females from damage I collected them very carefully in adapted film boxes with a gauze covered opening at each end. Thus the insects could hold on to the gauze and were well supplied with air. Usually I marked the specimens the following morning because their wings had hardened by that time. Codes of points and numbers in different colours were marked on the wings using a waterproof pen (Staedtler Lumicolor 313). Every evening I weighed all females and, beginning on the 6th of July, I also measured the maximum width of the 7th abdominal segment using a dissecting microscope (Zeiss Stemi SR, magnification 32 x). The balance was a Mettler PM 100 (precision: 1 mg). The specimens were transported in the film boxes and weighed in them. Their colour was noted daily. The temperature in the cage was measured daily using a minimax-thermometer.

To study the influence of food supply on the time of colour change, 58 freshly emerged females were caught and weighed on the 16th of July. The following morning they were put into two cages as described above, 29 in each cage. The females in cage 1 were fed every evening (except on the 7th day). Those in cage 2 were fed only once (6th day; 1/2 quantity). On the morning of the 8th day the experiment was terminated. All the damselflies which had survived were weighed, their abdominal width measured and their colour noted. Insects which had just started to change colour were counted as green females.

TIME OF COLOUR CHANGE

The time of colour change was variable and depended on several factors, e.g. temperature and food supply. Usually colour transformation took one or two days. The first day on which a change in colour could be observed is referred



Fig. 1. Day of colour change in female *I. pumilio* as function of emergence date: group (a), less well-fed individuals [n=13, f(x)=-0.251x + 12.294, r=0.899, p=0.0001]; - group (b), well-fed individuals [n=18, f(x)=-0.239x + 10.762, r=0.464, p=0.0524]. - The dot size corresponds to the respective number of individuals.

seven days, respectively, for it to occur. High mortality in both groups was caused mainly by spiders which regularly entered the cage, and partly especially in group (a) by cannibalism.

to as the day of colour change. In group (a) 14.1% of the damselflies survived until colour transformation. Their mean livespan was 5.6 days. Females in group (b) lived for a mean of 7.9 days, 58.1% of them have reached the time of colour change. In group (a) there was a considerable difference between females which emerged before the 12th day of the experiment (n=66) and those which emerged after the 23rd day (n=26). The former lived a mean of 6.3 days. 16.6% of them survived until colour change which took a long time (mean: 11 days, n=11). The latter lived a mean of only 3.8 days but the two females which survived until colour transformation (7.7%) took only six and Both groups showed a negative correlation with the date of emergence (Fig. 1), as well as with the sum of the maximum daily temperatures during the first six days of life (Gr. (a): r=0.777, p<0.005; Gr. (b): r=0.454, p=0.059). In both groups the time of colour change was more highly correlated with the date of emergence than with temperature. Because of the very short period when weight and abdominal width could be measured in group (b), the results are only tentative. The regression curves of the two groups run nearly parallel but group (b) is shifted downwards. Females in this group took less time to reach colour transformation than those in the other group. Group (a) took a mean of 10.3 (\pm 2.06) days and group (b) a mean of 7 (\pm 0.77) days (U=26.5, p<0.001).

In the field I caught orange females while they were sitting in a copulation wheel (n=10). Marked individually and put into an outdoor cage, eight of them changed or started to change colour to green the day after their capture. One of them I could not find and one female was still orange.

DEVELOPMENT OF WEIGHT AND ABDOMINAL WIDTH

Directly after emergence the weight of all females dropped significantly. After a short increase weight dropped again in less well-fed individuals (Fig. 2A) and then increased slowly. There was another drop in weight directly before colour transformation occurred. After this weight increased again and reached its maximum three days after the day of colour change. In contrast, the weight of wellfed females (Fig. 2B & C) increased continuously after the first drop. On the third and fourth day, weight gain was not significantly higher than on the first day, but on the fifth day it was so. The colour change occurred one or two days later. One day after colour change the weight reached a temporary maximum, and thereafter it started to vary. Weight development took a similar course in females which changed colour after six or seven days (Fig. 2B & C), but the rate of increase was greater especially between the fourth and fifth day in those which changed colour after six days. Abdominal width developed more or less in parallel to weight, but without the drop at the beginning.

Parameters	Cage 1	Cage 2	U	Р
Initial number	29	29		
Final number	11	5		
Initial mean weight (mg) (± SD)	23.4 (± 2.08)	22.9 (± 2.25)	376	n.s.
Final mean weight (mg) (± SD)	29.8 (± 4.13)	23.2 (± 1.30)	0	< 0.005
Final mean width of abdomen (mm) (± SD)	1.05 (± 0.05)	0.93 (± 0.45)	0	<0.005
Final body colour				
Orange	0	4		
Green	11	1		

Table 1

Influence of food supply on the development of immature females of Ischnura pumilio

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Colour change in female Ischnura pumilio



FOOD SUPPLY EXPERIMENT

The development of individuals was very different in the two cages. Initially there was no significant variation of weight (Tab. I). During the experiment mortality was high in both cages, 62% in cage 1 and 83% in cage 2. After the

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experiment, females in cage 1 were significantly heavier and had a significantly broader abdomen than those in cage 2 (Tab. I). The Fisher-Yates Test was used to measure the colour distribution. It showed a significantly higher number of green females in cage 1 than in cage 2 (Tab. I; p<0.01).

DISCUSSION

Maturation in dragonflies takes between two days and several weeks (e.g. CORBET, 1962). In the genus *lschnura* it takes between 4 and 14 days. The shortest times are reported for *l. verticalis* with 4.5 ± 0.4 days (FINCKE, 1987) or 4-7 days (GRIEVE, 1937), respectively. DUNKLE (1990) states that *l. posita* takes about 7 days until changing colour and PARR (1973) found a maturation time of 6, 7 and 8 days for the different colour variations of female *l. elegans* and 6.2 days for males. *l. gemina* takes about 7-10 days (GARRISON & HAFER-NIK, 1981) and HINNEKINT (1987) reports a maturation time of 14 days for male *l. elegans*.

The 6-12 days maturation time I found for I. pumilio fits well with the above data. S.A. Cham (pers. comm.) found a maturation time of 5-9 days for I. pumilio using a mark-recapture programme. When comparing species with each other, attention must be paid to the fact that all these values have been obtained using different methods and under different climatic conditions, since it is just such factors, and in particular the food supply, which have a great influence on maturation time, as this study has shown. Females in group (b) took a mean of 7 days until the time of colour change. This is similar to the time taken by I. posita. In contrast, females with a lack of food took a mean of 10 days, which is similar to the time taken by *I. gemina*. Thus, differences between species may disappear when the factors influencing maturation vary. Another indication for this is that HINNEKINT (1987), contrary to PARR (1973), found a maturation time of 14 days in male I. elegans. PARR (1973) used a mark-recapture programme, while HINNEKINT (1987) kept the damselflies in a cage (8 x 4 x 4 cm per 2 specimens) under laboratory conditions. He cut the wings at the nodus and fed the insects with live Drosophila melanogaster by forceps. I know from my own experience that damselflies often take their food unwillingly while held in the hand. HINNEKINT (1987) himself stated that individuals refusing food almost invariably died the next day. The other damselflies consumed an average of four flies per day, while those of LORD (1961, as cited by HINNEKINT, 1987), who kept individuals of the same species in outdoor cages, ate eight items of prey per day. He concluded that his specimens used less energy because they could not fly. Nevertheless, I suggest that the damselflies took such a long time for maturation because the treatment hampered their development. A dependency of maturation on food supply seems logical, but this study clearly shows that there must be at least one additional factor which influences maturation time. This factor is temperature which my experiments showed to be inversely proportional to the time taken until colour transformation. BUCHHOLTZ (1951) supposed that maturation will be shortened at higher temperatures and others have also reported temperature dependent maturation times (e.g. MILLER, 1987).

The fact that the time until colour change was more highly correlated with the date of emergence than with the temperature, indicates the importance of another factor, such as wind, rain, light or the day length. The later in the season a damselfly emerges, the faster it needs to reach maturity to ensure that it will be able to complete its lifecycle. It is true that the data were obtained long before the end of the flight period of *I. pumilio*, but frequent rainfalls and low temperatures made weather conditions extremely bad in the study area during the entire investigation period.

As far as I know, individual development of weight and width of abdomen have not been recorded in Odonata until now. Reports on the stage of maturity of the different colour forms in female Ischnura have been based only on dissections. KYLE (1961) and ZIMMERMANN (1973) who carried out dissections of female I. pumilio, never found an orange female with fertile eggs. ZIMMER-MANN (1973) also weighed several individuals of I. pumilio and found that green females were always heavier than orange ones. I have found the same in females caught in the field. The drop in weight immediately after emergence may be explained by a low rate of feeding. Of course, it is possible that this drop lasted a bit longer in individuals kept in cages than in those in the field because they were disturbed by catching and weighing. But in my experience, damselflies in the field spend the first day of life sitting in the vegetation to be better protected from predators. The drop in weight is followed by an increase in body weight and a build up of body substance. In females this involves, above all, the abdominal region and the ovaries, as ANHOLT et al. (1991) showed for I. verticalis and a number of other species. Therefore I have felt justified in using weight and width of abdomen as measures of sexual maturity. The relationship between sexual maturity and female colour change is clearly seen, because most females changed their colour shortly before reaching the first maximum. Variation in weight which occur later can be explained by oviposition and, above all, by the decrease in the number of females. Especially the death of lighter individuals gives the impression that weight increases later.

As this study shows, orange females start to copulate at the earliest shortly before colour transformation. So far as I know, orange females of *I. pumilio* have never been seen laying eggs. Thus, the copulations observed are either fictitious copulations or, more likely, the females are able to store the sperm until they are capable of laying eggs.

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