Effects of suppressed oviposition activity and flight muscle histolysis on food consumption and ovarian development in a wing-dimorphic cricket: an explanation for sporadic conclusions related to physiological trade-offs

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Abstract

The effects of deprivation of oviposition substrate on food consumption and egg production were compared between the long-winged (LW) and the short-winged (SW) morph of a cricket, Modicogryllus confirmatus, to determine how suppressed oviposition activity would influence these traits in each wing morph. Food consumption was greatly suppressed in females deprived of oviposition substrate (−OS) compared to those given access to it (+OS) during the 2-week feeding trial in the SW morph but not in the LW morph. Some LW females shed their hindwings and histolyzed the flight muscles. Such de-alated LW (DLW) morphs tended to consume more food than intact LW (ILW) morphs. In all morphs, ovarian weight was heavier under −OS conditions than under +OS conditions during the second week of adulthood, although the differences were greater in SW morphs than in ILW morphs. In DLW morphs in which flight muscle histolysis was induced by artificial de-alation at adult emergence, the temporal changes in ovarian weight were similar to those of SW morphs.

In SW morphs, food consumption was also significantly reduced when ovipositing females were deprived of oviposition substrate for 2 days compared to those allowed to oviposit continuously, but food consumption was not reduced in ILW or DLW morphs. SW females from which one ovary was extirpated at adult emergence, SW (−o), also showed a significant difference in food consumption when treated as above, indicating that food consumption was not determined simply by the number of ovarian eggs. The crop content was positively correlated to food consumption and smaller under −OS conditions than under +OS conditions.

These results indicate the possibility that some inconsistent results and conclusions discussed in recent studies, concerning the physiological trade-offs between flight capability and reproduction, were caused by the suppressed oviposition activity and failure to recognize the occurrence of flight muscle growth and histolysis in the test crickets. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Wing dimorphism; Physiological trade-off; Oviposition; Food consumption; Egg production; Flight muscle histolysis; Crickets

1. Introduction

Wing dimorphism is commonly found in various insect orders including Orthoptera, Dermaptera, Zoraptera, Psocoptera, Thysanoptera, Hemiptera, Homoptera, Coleoptera, Lepidoptera, Diptera and Hymenoptera (Johnson, 1969; Borror et al., 1976; Harrison, 1980; Din- gle, 1985; Roff, 1986; Fujisaki, 1994). In general, two morphologically distinct morphs, long-winged (LW) and short-winged (SW), occur and exhibit various physiological and behavioral differences. In some groups, apterous morphs are known, but in terms of the dispersal capability, they are equivalent to the SW morph. The LW morph normally has well-developed flight muscles and is regarded as a migratory morph. On the other hand, the SW morph with reduced wings and flight muscles...
cannot fly but reproduces earlier and in many species is more fecund than the LW morphs (Harrison, 1980; Dingle, 1985; Roff, 1986; Zera and Denno, 1997). The general consensus is that the absence of the large wings and functional flight musculature allows more energy to be devoted to reproduction and survival (Dingle, 1985). Therefore, it is reasonable to assume that there is an energetic cost involved in the development and maintenance of the flight capability in the migratory morph. This idea is based on an assumption that resources are limited within the organism, and thus trade-offs occur between different life-history traits (Calow 1978, 1979; Stearns, 1992). However, the presence of a correlation between wing morph and reproductive characteristics does not necessarily prove their causal relationship (Tanaka, 1993; Tanaka and Suzuki, 1998).

To address issues concerning flight capability in wing dimorphic insects, physiological studies are necessary. Mole and Zera (1993, 1994) have compared nutritional indices and flight muscle development profiles between LW and SW morphs of the crickets Gryllus rubens and G. firmus to identify potential physiological costs of flight. Their main findings are as follows. (1) There was no direct trade-off between flight muscle and ovarian growth. (2) There was no intermorph difference in food consumption in G. rubens, but LW morphs ate more food than SW morphs in G. firmus. (3) LW morphs converted a lower proportion of assimilated nutrients into biomass (i.e. mainly the ovary) than did SW morphs. (4) Ovarian growth was greater in SW morphs than in LW morphs in G. rubens, but no difference was found in G. firmus. (5) No intermorph difference was found in the number of eggs laid in either species, in contrast to previous results obtained for these (Roff, 1984; Zera and Rankin, 1989) and other species (Tanaka 1976, 1993). These results, together with those reported by Zera et al. (1994), provided the basis for the trade-off theories discussed in more recent articles by Zera and his colleagues (Zera and Denno, 1997; Zera et al., 1998).

It seems surprising that the closely related species of G. rubens and G. firmus show such physiological inconsistency. In fact, some of the findings related to fecundity and flight muscle development contradict earlier ones with the same and other species, as pointed out by Crno krah and Roff (1998), Tanaka and Suzuki (1998) and Tanaka et al. (1999). In crickets, it has been known that ovarian development and food consumption are influenced by various factors such as temperature, mating, and the presence or absence of oviposition substrate (Loher and Edson, 1973; Clifford and Woodring, 1986; Loher et al., 1987; Renucci et al., 1990). One of the consistent results in the studies by Mole and Zera (1993, 1994)) is the unusually low ovipositing activity in spite of the fact that all females were mated; in the first two weeks of adult life, G. firmus females produced only 83 eggs on average and G. rubens females only 20 eggs. As pointed out by Tanaka and Suzuki (1998), this is in marked contrast to >400 for G. firmus reported by Roff (1990) and >250 reported for G. rubens by Zera and Rankin (1989). In the present study, I conducted experiments on egg retention to mimic this low egg-laying in a cricket. In a more recent study, Zera et al. (1994) reported intermorph differences in lipid and carbohydrate contents in G. firmus, but all test females were not allowed to lay eggs during the entire period of the experiment. The possible impact of suppressed ovipositing activity on various physiological traits as well as on trade-offs was pointed out by Tanaka and Suzuki (1998) and Tanaka et al. (1999), but no study has been designed to examine these specific problems.

In the present study, I investigated the effects of suppression of ovipositing activity on food consumption, ovarian development and body weight in female adults of a wing dimorphic cricket, Modicogryllus confirmatus. Because food consumption and ovarian weight are known to be influenced by the development and histolysis of the flight muscle in LW morphs of this species (Tanaka, 1993; Tanaka et al., 1999), a group of LW morphs in which flight muscle histolysis was induced by artificial de-alation at adult emergence (Tanaka, 1994a) was included to determine the role of flight muscle histolysis in the control of food consumption under stressful conditions. In crickets, flight muscle histolysis is common in LW morphs, and in several species LW females with histolyzed flight muscles develop their ovaries as rapidly as SW females (Tanaka 1976, 1986; Tanaka, 1994a,b; Zera et al., 1997). The results of these observations are reported in this paper. They strongly indicate that suppression of ovipositing activity and failure to recognize the occurrence of flight muscle histolysis could (1) partly explain the above-mentioned sporadic results reported by Zera and his colleagues (Mole and Zera 1993, 1994; Zera and Mole, 1994; Zera et al., 1998), and (2) call into question the validity of the biochemical comparison between wing morphs in crickets deprived of oviposition substrate (Zera et al., 1994). Hence some of the generalizations made by Zera and Mole (1994), Zera and Denno (1997) and Zera et al. (1998) may also need re-evaluation.

2. Materials and methods

The crickets used were reared at 30°C and 16 h light/8 h dark photoperiod, as previously described (Tanaka, 1993; Suzuki and Tanaka, 1996). They were fed dry pellets (Insect Feed, Oriental Yeast Co., Tokyo), both as nymphs and adults.

In the first experiment, I examined the effect of oviposition substrate on food consumption and body weight in SW and LW females. Newly emerged female adults of each wing morph were weighed and divided into two
groups. They were individually held in small plastic containers (diameter 10 cm; height 5 cm) each having a lid with a wire-mesh-covered hole (diameter 3 cm). All individuals were handled in the same way during the first 3 days. In each container, 100 mg of food and a large water vial (diameter 3 cm; length 6 cm) plugged with cotton-wool were provided during the first 3 days. Care was taken to prevent the food from getting wet by using the method described previously (Tanaka et al., 1999). Adults of this cricket do not lay eggs during the first 3 days (Tanaka, 1993). On day 3, the food left was weighed and each cricket was given a chance to mate with a sexually mature male in another container where no food or water was provided. Most individuals mated within 10 min and all mated females with a spermatophore were returned to the original container after a 1-h mating trial and given 200 mg of food. A few females did not mate and they were discarded.

In each wing-morph group, one subgroup of females was constantly provided with a large water vial plugged with cotton-wool, and the other subgroup with a small water vial (diameter 1 cm; length 5 cm) plugged with cotton-wool. Crickets laid eggs into the cotton plug of the large vial, but failed to do so when given the small vials. However, a few crickets of the latter subgroup managed to lay eggs and some deposited eggs on the dry floor of the container. These individuals were excluded from the analysis. All females were weighed and given more food every 2 or 3 days until day 14. Some LW females shed the hindwings during the experimental period. Because such de-alated LW individuals are known to histolyze the flight muscle and to start rapid reproduction (Tanaka, 1993), they were separated as de-alated LW (DLW) from the intact LW (ILW) adults. In this experiment, crickets were always given plenty of food and fed ad libitum. At the end of the experiment, the total amount of food consumed was calculated.

In a second experiment to determine ovarian development in crickets deprived of oviposition substrate (−OS) and those given access to it (+OS), newly emerged male and female adults were kept together in a large container (20×35×26 cm) where they were fed ad libitum. As in the above experiment, some adults were allowed to lay eggs freely and the others deprived of oviposition substrate. In addition to the SW and ILW groups, another group, DLW, in which the hindwings were artificially removed from ILW females at adult emergence was included to test the effect of flight muscle histolysis on body weight and ovarian development. Crickets were sampled randomly from each group on days 7, 10, and 14, individually put into an air-tight tube (Eppendorf, volume 1.5 ml), and stored at −20°C. The frozen crickets were thawed within a week at room temperature, weighed, and their ovaries were dissected out to determine the mass by using a Mettler AT 201 balance.

In a third experiment, both male and female adults were reared as a group in a large container, and 4-day-old females were individually kept in small containers and given plenty of food and a large water vial with a cotton plug. On the next day, those females which had deposited eggs were individually transferred to new containers each holding 100 mg of food and a small or a large water vial. In this case, crickets of the same wing morph were divided into two subgroups of equal mean body weight to exclude the effect of body size. In addition to the SW and ILW groups, another group, SW (−o), in which one ovary was removed at adult emergence from SW females, was included to examine the effect of ovarian mass on food consumption and other developmental traits. For extirpation of ovaries, newly emerged females were kept on ice for about 20 min and the right ovary was pulled out of the body with a fine forceps through a small incision made between the fourth and fifth abdominal tergites. At the end of the 2-day confinement, the food left was weighed to determine the food consumption rate, and the eggs deposited into the cotton plugs were counted for the +OS subgroups. Among the ILW group, some individuals shed their hindwings during the experiment, and such individuals were separated as DLW morphs. All crickets were stored at −20°C as above, and dissected later to determine weights of the whole body, crop content and ovaries. The chironionated oocytes or eggs in the ovaries were also counted. The dry weight of the dorsolongitudinal flight muscle was determined for ILW and LDW individuals according to the method of Tanaka (1993). The data were analyzed by ANOVA followed by a Games–Howell test or a t-test (Statview).

3. Results

3.1. Effects of deprivation of oviposition substrate in SW and ILW

Deposition of oviposition substrate reduced food consumption rate significantly in SW females (Fig. 1A). Food consumption rates were similar between the first and second weeks under +OS conditions, but the rate was reduced by about 50% in the second week under −OS conditions (t = 6.00; df = 32; P < 0.001). In ILW females, on the other hand, no such difference was found (Fig. 1B). SW crickets (mean±SD, 373.6±83.0 mg; N=20) ate significantly more food during the 2 weeks than ILW ones (281.7±89.6 mg; N=14; P < 0.05) under +OS conditions, but no significant difference occurred under −OS conditions (mean±SD=212.6±34.8 mg for SW, N=14; and 281.7±89.6 mg for ILW, N=7). These results demonstrated that food consumption was strongly suppressed by deprivation of oviposition substrate in SW females, but not in ILW ones, although the mean values...
Fig. 1. Effect of deprivation of oviposition substrate on mean food consumption during the first 2 weeks after adult emergence in SW (A) and ILW (B) females of *M. confirmatus* at 30°C. Some LW females underwent natural de-alation during the experiment and separated as DLW morphs (C). Open and closed histograms indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively. Bars on histograms indicate one SD. An asterisk indicates a significant difference between the two subgroups by a t-test (*P*<0.05). *N*=6–20.

for −OS crickets were consistently smaller than those for +OS crickets in both cases.

As mentioned, some ILW shed their hindwings during the experimental period. This de-alation occurred in 30% of individuals under +OS conditions and 43.8% under −OS conditions. Although the timing of de-alation varied from day 2 to day 10 among individuals (mean±SD=4.3±2.7 days and 4.3±2.5 days in +OS and −OS DLW females, respectively), their flight muscles had been completely histolyzed by the end of the experimental period. Under both +OS and −OS conditions, DLW females showed changes in food consumption rate almost equivalent to those for SW females. The differences between SW and DLW females under either +OS or −OS conditions were all insignificant (*P*>0.05) except for the comparison during the second week under −OS conditions (*t*=3.385; df=19; *P*<0.05). Although the differences were not significant, the mean food consumption rate for DLW females was consistently greater than that for ILW females under either +OS or −OS conditions for each feeding period. It should be noted that −OS ILW females consumed significantly less food than +OS DLW females during both the first (*t*=0.457; df=11; *P*<0.01) and second (*t*=5.910; df=11; *P*<0.01) weeks, indicating that −OS ILW crickets would increase rates of food consumption greatly at any time if they shed their hindwings and oviposition substrate becomes available.

In the above experiment, the mean and SD values of initial body weight determined at adult emergence (about 6 h post-emergence) were 137.9±11.6 (*N*=20) and 132.1±10.0 (*N*=14) for the SW subgroups given access to oviposition substrate (+OS) and no access (−OS), respectively. The corresponding figures for the ILW counterparts were 136.2±11.4 (*N*=14) and 134.9±16.8 (*N*=9). No significant difference was found in initial body weight between the four subgroups, indicating no effect of body size on food consumption and subsequent growth.

Fig. 2 shows the changes in body weight on day 7 and later. In SW morphs, body weight tended to be heavier under −OS conditions than under +OS conditions; the differences were small but all significant (*t*=3.91, 2.13 and 3.72 on days 7, 10 and 14, respectively; *P*<0.05 in all cases). In LW morphs, deprivation of oviposition substrate did not influence body weight significantly. On day 14, SW crickets were significantly heavier than ILW ones irrespective of the presence or absence of oviposition substrate (Games–Howell test; *P*<0.05), but not different from DLW crickets in body weight. In all groups, body weight increased during the first week, but showed no further change thereafter (ANOVA, *P*<0.05 each).

Fig. 3 compares the ovarian weight as a percent of the whole body mass between +OS and −OS crickets reared in another series of experiments. The ovaries of −OS crickets were significantly heavier than ILW ones irrespective of the presence or absence of oviposition substrate (Games–Howell test; *P*<0.05), but not different from DLW crickets in body weight. In all groups, body weight increased during the first week, but showed no further change thereafter (ANOVA, *P*>0.05 each).

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Fig. 2. Effect of deprivation of oviposition substrate on mean wet body weight during the first 2 weeks after adult emergence in SW (A) and ILW (B) females of *M. confirmatus* at 30°C. Some LW females underwent natural de-alation during the experiment and separated as DLW morphs (C). Open and closed histograms indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively. Bars on histograms indicate one SD. An asterisk indicates a significant difference between the two OS subgroups by a *t*-test (*P* < 0.05). *N* = 6–20.

Fig. 3. Effect of deprivation of oviposition substrate on mean ovarian weight as a percent of wet body weight in SW (A), ILW (B) and DLW (C) female adults of *M. confirmatus* at 30°C. All DLW individuals are those which were artificially de-alated at adult emergence. Open and closed histograms indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively. Bars on histograms indicate one SD. An asterisk indicates a significant difference between the two subgroups by a *t*-test (*P* < 0.05). *N* = 5–13.

ies represented only 12.0–13.6% under +OS conditions and 17.2–20.8% under −OS conditions. The differences between SW and ILW morphs in ovarian development were remarkable, and all were significant under either +OS or −OS conditions (*t*-test; *P* < 0.05). The DLW morphs in which the flight muscles had been histolyzed completely before day 7 were similar in results to the SW morphs. Deprivation of oviposition substrate apparently caused crickets to accumulate eggs in the ovaries, and flight muscle histolysis influenced ovarian development in LW crickets.

3.2. Effects of 2-day deprivation of oviposition substrate

To observe how ovipositing females would respond to a short period of stress due to suppressed oviposition activity, three groups of crickets, ILW, SW, and SW (−o) with one ovary extirpated at adult emergence, were confined individually in containers under +OS or −OS conditions for 2 days. Extirpation of one ovary was undertaken to examine the hypothesis that the enlarged ovaries caused by failure to lay eggs would result directly in a reduction in food consumption. As shown in Fig. 4A, deprivation of oviposition substrate caused a significant decrease in food consumption of SW females and SW (−o). Under +OS conditions, SW females deposited almost exactly twice as many as SW (−o) ones (Fig. 4B). These results may indicate that these females had been producing eggs at a maximal rate under +OS conditions, and the difference in egg production was simply related to the number of ovarioles. When the total number of eggs including deposited eggs and ovarian eggs was compared (Fig. 4C), however, deprivation of oviposition substrate did not cause any significant difference in either group, and intact SW females had twice as many eggs as SW (−o) females under both conditions. These results indicated that depri-
Fig. 4. Effects of 2-day deprivation of oviposition substrate from 5-day-old ovipositing female adults on food consumption (A), numbers of deposited eggs (B), total numbers of eggs including deposited and ovarian eggs (C) and total (reproductive) output (D) in *M. confirmatus* at 30°C. Total reproductive output means the combined weight of the deposited eggs and ovaries. In addition to SW, ILW and DLW groups, an SW (−O) group in which one ovary had been extirpated at adult emergence from SW females was also included. Open and closed histograms indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively. Bars on histograms indicate one SD. An asterisk indicates a significant difference between the two subgroups by a *t*-test (*P*, 0.05). Different letters in or on histograms indicate a significant difference by a Games–Howell test (*P*, 0.05) under respective conditions except for (C) in which the data under +OS and −OS conditions were not significantly different from each other and the combined data were compared between groups. *N*=5–18.

Vivication of oviposition substrate reduced food consumption significantly even in crickets with only one ovary, and extirpation of an ovary caused egg production to decrease by roughly 50%.

A slightly different result was obtained for ILW. As mentioned, some ILW shed their hindwings spontaneously during the 2-day experimental period, and they were separately analyzed as a DLW group. At dissections, the flight muscles were examined for each individual: all ILW individuals had well-developed pinkish muscles (mean dry weight±SD, 2014.4±148.8 µg; range, 1730–2230 µg; *N*=23), whereas DLW ones had a mixture of well-developed and histolyzing muscles (mean dry weight±SD, 1368.1±438.5 µg; range, 800–2090 µg; *N*=16), the difference in mean being highly significant (*t*=6.577; df=37; *P*<0.001). The histolyzing muscles were either pink, yellow, or whitish in color, indicating that flight muscle histolysis started at different times in different individuals. In either ILW or DLW groups, mean food consumption was greater under +OS conditions than under −OS conditions, but the differences were not significant at the 5% level (Fig. 4A). Deprivation of oviposition substrate did not cause a significant difference in total egg production, as in SW morphs.

In Fig. 4, the difference in food consumption caused by deprivation of oviposition substrate was not reflected in total egg production in the SW groups: mean total egg production was similar (*P*>0.05) or even slightly larger in the −OS females that had consumed less food during the 2-day period than their +OS counterparts. Two factors appeared to be implicated in this seemingly paradoxical phenomenon. One was the crop content, which had a significant positive correlation with food consumption overall: the larger the amount of food consumed the larger the crop content (Fig. 5), though a significant correlation was observed only under +OS conditions (*r*=0.358; *N*=51; *P*<0.01). However, the difference in crop content between +OS and −OS individuals amounted only to about 7 mg in wet weight (Fig. 6), which was less than 3 mg in dry weight because crop contents had 66.3% of water on average (S. Tanaka, unpublished data). ANOVA indicated a significant difference in crop content among the four groups under...
Fig. 6. Effect of 2-day deprivation of oviposition substrate from 5-day-old ovipositing female adults on mean crop content in SW, SW (−o), ILW and DLW groups of *M. confirmatus* at 30°C. For explanation of groups, see Fig. 4. Crop contents were determined on day 7. Open and closed histograms indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively. Bars on histograms indicate one SD. An asterisk indicates a significant difference between the two subgroups by a *t*-test (*P*<0.05).

−OS conditions (*F*=3.239; df=3, 44; *P*<0.05), but no significant difference under +OS conditions. Fig. 7A shows that SW females laying eggs under +OS conditions had a smaller ovarian mass than those under −OS conditions at the end of the experiment, but the ovaries of the former contained many developing oocytes, which represented 76.8% of the total ovarian mass. Under −OS conditions, the ovaries of SW females under −OS conditions were probably close to the maximal size possible, judging from the body weight changes under −OS conditions in Fig. 3. These results indicated that most nutrients assimilated by feeding were used for maturing eggs under −OS conditions, whereas that was allocated to development of both eggs and new oocytes under +OS conditions. When the total reproductive output including all eggs produced and developing oocytes was calculated (Fig. 8), it was greater for +OS females than for −OS females in the SW group, and positively correlated with food consumption. Fig. 8 illustrates the relationships between food consumption during days 5–7 and total reproductive output for all individuals tested in the above experiment. The two variables showed a highly significant positive correlation (*r*=0.769; df=97; *P*<0.001), which indicated that, in general, the rate of food consumption which was influenced by various factors affected total reproductive performance directly.

In SW (−o) females, however, the difference between the +OS and −OS groups was not significant either in crop content (Fig. 6) or in the total mass of developing oocytes (Fig. 7A). These individuals had only one ovary. Thus, it is possible that the ovary was developing eggs and oocytes at a maximal rate under both conditions so that the reduced food consumption in −OS females did not affect the total reproductive output (Fig. 4D). The excessive food ingested by +OS individuals was probably not assimilated, because their net body weight (228.0±32.5 mg; *N*=5), determined after removal of the crop content and the ovary from the whole body, was equivalent to that for −OS individuals (222.35±20.57; *N*=6; *P*>0.05).

In the ILW group in which food consumption rate was

Fig. 7. Effect of 2-day deprivation of oviposition substrate from 5-day-old ovipositing female adults on mean ovarian weight in *M. confirmatus* at 30°C. For explanation of groups, see Fig. 4. For each group, the left histograms indicate females kept with oviposition substrate (+OS) as controls and the right those without it (−OS). The closed area indicates ovarian eggs and the hatched area developing oocytes. The negligible weight of the ovarian tissue (ca. 2 mg) was included as a part of developing oocytes. Bars on histograms indicate one SD. *N*=5–18. An asterisk indicates a significant difference in mean ovarian weight between the two subgroups by a *t*-test (*P*<0.05). Different letters in or next to histograms indicate a significant difference by a Games–Howell test (*P*<0.05) under the respective conditions.

Fig. 8. Total reproductive output (deposited eggs plus ovaries) plotted against the amount of food consumed during 2-day deprivation of oviposition substrate from 5-day-old ovipositing female adults in *M. confirmatus* at 30°C. Data for all groups given in Fig. 4 are pooled (*r*=0.402; df=97; *P*<0.001). Open and closed symbols indicate females kept with (+OS, control) and without oviposition substrate (−OS), respectively.
not influenced by deprivation of oviposition substrate, no significant difference was observed in either the total mass of developing oocytes (Fig. 7) or the total reproductive output (Fig. 4D). In the DLW group, no significant difference was found in total reproductive output between +OS and −OS conditions, but +OS adults had more developing oocytes than −OS adults. It should be noted that DLW adults consumed more food and had more total reproductive output than ILW ones (Figs. 4A and D). At dissections, most ILW individuals (91%) had well-developed, pinkish flight muscles, whereas the pinkish flight muscles were found in only 18.8% of DLW individuals and the rest had yellow or whitish muscles, indicating that the flight muscles were histolysing in the latter. The mean dry mass of the dorsolongitudinal muscle was 2024.4 μg (±148.8, SD; N=23) for the ILW group and 1368.1 μg (±438.5, SD; N=16) for the DLM group, and the difference between the two groups was highly significant (t=6.577; df=37; P<0.0001). These results revealed that the onset of flight muscle histolysis accompanied by de-alation increased food consumption and total reproductive output even under −OS conditions.

4. Discussion

The present results demonstrated that food consumption and reproductive development were greatly influenced by the presence or absence of oviposition substrate in mated crickets. Although these findings are not new and have been reported in other cricket species such as Acheta domestica (Renucci et al., 1990) and T. commodus (Loher and Edson, 1973; Clifford and Woodring, 1986; Loher et al., 1987), the importance of oviposition activity in the control of food consumption rate and reproductive development has not been compared between wing morphs or between the flight-capable LW group and the flightless group including SW and DLW. The present study was undertaken in order to examine the possibility that suppressed oviposition activity could explain some of the recently published conflicting results and conclusions related to physiological trade-off reported by Zera and his colleagues (Mole and Zera 1993, 1994; Zera and Mole, 1997; Zera et al. 1994, 1997), as outlined in Section 1.

4.1. Flight muscle growth and histolysis

Before going into further detail, we need to clarify some specific problems that are important in terms of physiological trade-off between flight capability and reproduction in crickets. In several species that my colleagues and I have studied, the flight muscles in the LW morph were well developed at adult emergence, but they all increased in mass during the early stage of adulthood. Examples include Allonemobius fasciatus (Tanaka, 1986), A. socius (S. Tanaka, unpublished), Velarifictorus parvus (Tanaka, 1991), Gryllus bimaculatus (Gomi et al., 1995), Teleogryllus derelictus (S. Tanaka, S. Arai and K. Nakamura, unpublished), Diademobius mikado, D. nigrofasciipes and Pteronomobius nitidus (S. Tanaka, unpublished). In G. rubens and G. firmus, Zera and colleagues (Mole and Zera 1993, 1994; Zera and Mole, 1994) reported no changes in mass of the flight muscles in the LW morph during the first 2 weeks after adult emergence. Based on this finding, they concluded that there is no direct trade-off between flight muscle and ovarian growth in adults (Zera and Mole, 1994; Zera and Denno, 1997). However, because they compared only mean flight muscle masses in their studies, there is the possibility that some individuals grew flight muscles and others histolysed them, resulting in mean values that did not reflect any significant change during the early stage of adulthood, as pointed out by Tanaka and Suzuki (1998). In G. firmus, the occurrence of flight muscle histolysis was first reported by Roff (1989) and recently reconfirmed by Zera et al. (1997, 1998), but no muscle growth was reported. In our preliminary observations with G. rubens from Tennessee in North America (S. Tanaka and T. Arai, unpublished), LW adults showed a substantial increase in flight muscle mass during the first 5 days in both sexes, as in M. confirmatus (Tanaka, 1993), but the details will be published elsewhere. In M. confirmatus, the presence of a direct trade-off between the flight muscles and reproductive organs has been demonstrated by controlling the food ration of SW, ILW and DLW morphs both in females (Tanaka, 1993) and males (Tanaka, 1999).

Maintenance of functional flight muscles is metabolically expensive (De Kort, 1969). There is no doubt that the massive pink flight muscles of the ILW morph require more energy for the maintenance of the tissue than the poorly developed whitish flight muscles of the SW morph (Tanaka and Suzuki, 1998). Therefore, it is reasonable to assume that the maintenance of functional muscles in the ILW morph is also a cost of flight capability (Tanaka, 1993). In fact, Zera et al. (1998) observed a significant difference in respiration rate of flight muscles in vitro between the two wing morphs of G. firmus. However, it appears that such a difference is rather insignificant relative to the metabolic rate of the whole body in crickets (Tanaka and Suzuki, 1998) and, in M. confirmatus, measurements of respiration rate of the whole body have revealed no significant difference either between the two wing morphs or between ILW and DLW male adults (Tanaka, 1999).

4.2. Food consumption and oviposition activity

In female crickets, food consumption rate is different between the two wing morphs in M. confirmatus
development influenced food consumption greatly. For example, it was reduced by about 25% in SW females during the first week if they were not allowed to lay eggs. The reduction in food consumption due to deprivation of oviposition substrate (−OS) was further pronounced in the second week, about 60% (Fig. 1). This reduced food consumption is correlated with the enlarged ovaries. Most females kept their eggs in the ovaries under −OS conditions and their ovarian weight increased to a high level during the first week and then remained almost the same thereafter (>30% of wet body weight relative to <20% under +OS conditions, Fig. 3).

A few SW females deposited eggs on the dry floor during the observations, and they increased food consumption immediately, though the data for these individuals are not presented in this paper. In ILW females, food consumption was not influenced significantly by deprivation of oviposition substrate, probably because their ovarian development is delayed compared to the SW morphs as demonstrated in this (Fig. 3; Tanaka, 1993; Tanaka and Suzuki, 1998) and other crickets, including Dianemobius mikado (Tanaka, 1976), Allonemobius fasciatus (Tanaka, 1986) and G. firmus (Zera et al., 1997).

Even under +OS conditions, the food consumption rate of M. confirmatus was smaller in the ILW morph than in the SW morph, conforming to the previous results (Tanaka, 1993; Tanaka et al., 1999). Some ILW females underwent de-alation and such individuals (DLW) increased food consumption both under +OS and −OS conditions. De-alation is accompanied by flight muscle histolysis and rapid egg production in many crickets and is thought to occur as a result of physiological change from the migratory to the reproductive phase (Tanaka, 1991; Tanaka, 1994a). In the first experiment (Fig. 1), no significant difference was obtained between +OS and −OS females in the DLW groups, probably because the time of de-alation varied greatly among individuals.

In the study by Mole and Zera (1993), oviposition activity was greatly suppressed in G. rubens during the entire experimental period of 2 weeks, but some females deposited eggs and in fact some appeared to have undergone flight muscle histolysis according to a recent article citing the results of this experiment (Zera et al., 1998). If this is true, the conclusions drawn by Mole and Zera (1993) and Zera and Mole (1994) should be reconsidered by taking the oviposition schedule and the timing and incidence of flight muscle histolysis into account. In G. firmus, the occurrence of flight muscle histolysis was recently found to be correlated with rapid ovarian development in LW females (Zera et al., 1997), as commonly observed in other crickets (Tanaka 1976, 1986; Roff, 1989; Gomi et al., 1995).

In M. confirmatus, both oviposition and flight muscle development influenced food consumption greatly. For example, food consumption was significantly greater in +OS DLW females than in −OS ILW females during the second week (Fig. 1C). This result implies that total food consumption during the 2 weeks will be increased substantially if ILW females that have not deposited eggs during the first week undergo flight muscle histolysis and start laying eggs in the second week, and also indicates that the oviposition schedule is another important factor influencing food consumption. A dramatic impact of de-alation on food consumption was also observed when ovipositing ILW underwent de-alation under +OS conditions (Fig. 7A). In other words, in M. confirmatus the total food consumption measured over a long period of 2 weeks can be similar between the two wing morphs or greater in one morph or the other depending upon these factors. It is obvious that the data without considering these factors, such as those reported by Mole and Zera (1993, 1994), are difficult to interpret and provide no meaningful information in terms of physiological trade-offs between flight capability and reproduction. Likewise, the biochemical comparison between the two wing morphs made for G. firmus (Zera et al., 1994) did not consider the status of flight muscles in LW morphs and all crickets were not allowed to lay eggs for as long as 12 days.

Zera et al. (1998) obtained a negative correlation between gut content and ovarian size in G. assimilis and suggested the presence of a “third party” trade-off between egg production and the volume of food that can be processed by the digestive system. However, this interpretation seems difficult to reconcile with the fact that no difference in food consumption or assimilation was found between the flight-capable and flightless individuals in this and other species, e.g. G. rubens (Mole and Zera, 1993), unless the digestive system is assumed to be more efficient when it is smaller. In M. confirmatus, a similar negative correlation was found between the crop content and ovarian weight when ovipositing females were deprived of oviposition substrate for 2 days in the present study, but the correlation was rather weak ($r=-0.88; N=48; P<0.05; data not shown$) because many individuals had small ovaries and small crop content. Under +OS conditions, on the other hand, the ovarian weight included many developing oocytes and Zera et al. (1994) did not consider the status of flight muscles in LW morphs and all crickets were not allowed to lay eggs for as long as 12 days.

4.3. Egg production and total reproductive output

In both the studies by Mole and Zera (1993, 1994), nutritional indices were compared between morphs at the end of the 2-week feeding trials without addition of deposited eggs, though inclusion of the masses of deposited eggs had no effect on the results of these analyses except for ECD (efficiency of conversion of digested food to body matter) values (see Zera and Mole, 1994). The technical problems associated with the application of nutritional indices to crickets have already been pointed out (Tanaka and Suzuki, 1998; Zera et al., 1998). The main purpose of the Mole and Zera experiments was to compare the food consumption, food assimilation and reproductive output (ovarian weight) between the two wing morphs. In G. firmus, ovarian growth reached a maximum in 5 days and no further growth occurred in the rest of the period in SW females (Mole and Zera, 1994). This is the pattern typically observed for SW females of M. confirmatus when food consumption was greatly suppressed under −OS conditions (Fig. 4), and the size of ovaries measured in such individuals probably indicated the maximum size of ovaries that crickets can hold in the body when oviposition is suppressed. In crickets under −OS conditions, the ovaries were enlarged and occupied almost exclusively by eggs, and few developing oocytes were found (Fig. 8), indicating a cessation of egg production. In G. rubens, the heavier ovarian weight at the end of 2-week feeding trials was found in SW females vs. LW females (Mole and Zera, 1993), and this result has often been cited as evidence for a greater reproductive potential in the SW morph than in the LW morph (Zera and Mole, 1994; Zera and Denno, 1997). In G. firmus, on the other hand, a comparison after the 2-week feeding trials indicated no difference in body weight between the two wing morphs, and increased food consumption by LW morphs was suggested to obviate the potential flight–fecundity trade–off (Mole and Zera, 1994; Zera and Denno, 1997).

Table 1 summarizes data showing how greatly the inhibition of oviposition activity influences the total egg production during the first 2-week period of adulthood in M. confirmatus. The mean total egg production estimated from the ovarian weight on day 14 was 249 eggs for SW females under −OS conditions, whereas the mean number of eggs deposited during the 2-week period by SW females under +OS conditions was 638 eggs. If the number of ovarian eggs estimated from the ovarian weight on day 14 is added, the total egg production for ovipositing SW females would amount to more than 750 eggs on average. The total egg production during the 2-week period under −OS conditions is three times less than that produced under +OS conditions in both SW and ILW adults. In view of such striking influences of the suppression of oviposition activity on egg production in this species and A. domesticus (Loher et al., 1987; Renucci et al., 1990), it is highly likely that a similar phenomenon also occurs in other crickets including G. rubens and G. firmus. If this is the case, the intermorph comparisons of ovarian mass or body weight at the end of the 2-week feeding trials in crickets that had laid only a small number of eggs (Mole and Zera 1993, 1994) would not have shown the potential reproductive capacity in either wing morph.

Although total reproductive output including deposited eggs, ovarian eggs and developing oocytes showed a highly significant positive correlation to the amount of food consumed (Fig. 8), food consumption rate was not reflected directly in total reproductive output in every case. In SW females from which one ovary had been extirpated at adult emergence, food consumption rate was significantly smaller when oviposition was prohibited for 2 days than when it was allowed continuously. However, this difference was not reflected in total reproductive output (Fig. 7). This result may indicate that oviposition activity influences the efficiency of food assimilation or digestibility.

4.4. Conclusion

Physiological trade-offs between flight capability and reproduction have been studied mainly with several species of crickets. As outlined above, recent studies reported some sporadic conclusions regarding intermorph differences in fecundity and food consumption in those species. It has been suggested that suppressed oviposition activity and failure to recognize the occurrence of flight muscle growth and histolysis could be the cause for such sporadic results and conclusions (Tanaka and Suzuki, 1998). In the present study, this hypothesis was tested by controlling oviposition activity and flight muscle development in M. confirmatus. As a result, evidence was obtained to indicate that both food consumption and fecundity can be similar between the two wing morphs or greater in one morph or the other.
depending upon these factors. Therefore, the present study strongly supports the above hypothesis, and also indicates that egg production measured for virgin or mated crickets without oviposition substrate available until far beyond the pre-ovipositional period is likely to be underestimated, although the conclusions drawn may not always be affected (e.g. Tanaka, 1986). For flight-fecundity trade-off studies, it is thus important to design experiments to avoid this problem. Intermorph comparisons of crickets reared without oviposition substrate should be limited to a short period of early adulthood.

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References

Tanaka, S., 1994b. Endocrine control of ovarian development and flight muscle histolysis in a wing dimorphic cricket, Modicogryllus confir-

Zera, A.J., Mole, S., Rokke, K., 1994. Lipid, carbohydrate and nitrogen...
content of long-winged and short-winged *Gryllus firmus*: implications for the physiological cost of flight capability. Journal of Insect Physiology 40, 1037–1044.
