Effects of starvation and mating status on the activity of the flea beetle, *Phyllotreta nemorum* (Coleoptera: Chrysomelidae)

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Key words. Chrysomelidae, *Phyllotreta nemorum*, female movement, flight activity, mating status, starvation

Abstract. Flea beetles are characterized by their tendency to jump. They can also fly. First, the effects of starvation on flight activity in the flea beetle, *Phyllotreta nemorum* L. (Coleoptera: Chrysomelidae) were determined. After starving *P. nemorum* for five days a greater number of individuals of both sexes flew than when fed continuously for the same period. In addition, the effect of the mating status of females of *P. nemorum* on their movement was determined. Mated females were more active than virgin females. It is concluded that starvation stimulates the flight activity of both sexes and mating activates the movement of females of *P. nemorum*.

INTRODUCTION

The flight activity of insects is affected by various factors, such as feeding condition. In some, but not all species, starvation stimulates flight activity (e.g., Elsey, 1974; Ferro et al., 1999; Moriya & Hiroyoshi, 1998). Jumping behaviour is characteristic of flea beetles and is well studied (e.g., Furth et al., 1983; Furth & Suzuki 1990, 1992). Although some species of flea beetle are apterous (e.g., Levesque & Levesque, 1998), others, including Phyllotreta nemorum L. (Coleoptera: Chrysomelidae), can fly (e.g., Lamb, 1983). In the laboratory flight is rarely observed, which raises the question – what conditions stimulate flight activity in flea beetles? Although the kinematics of the flight behaviour of flea beetles has been studied (e.g., Brackenbury & Wang, 1995) it is unclear what kind of physiological factors facilitate, or induce them to fly. Here, the effects of starvation on flight activity in *P. nemorum* are determined.

In addition, mating status often affects female movement; virgin females remain in areas where conspecific males are more likely to occur, whereas mated females do not (e.g., Julian & Gronenberg, 2002; Petersson & Sivinski, 2003). Thus, mating status may affect female movement (e.g., Shirai et al., 1998; Ferro et al., 1999). The effects of mating status on female movement in *P. nemorum* are reported here.

MATERIAL AND METHODS

Flea beetles

Flea beetle, *Phyllotreta nemorum* L. (Coleoptera: Chrysomelidae: Alticinae), is oligophagous feeding on a small number of plant species within the family Brassicaceae (Nielsen, 1977). Individuals of *Phyllotreta nemorum* were collected as larvae at Kvaerkeby and Try, Denmark in the summer of 2007 and reared on radish plants, *Raphanus sativus* L. (Brassicales: Brassicaceae), in a climate room (24 ± 5°C, 18L: 6D) at

Wageningen University, the Netherlands. The larvae pupated in the soil and the adults that emerged from the soil were used in the following experiments. *P. nemorum* does not mate immediately after emergence (K. Vermeer, pers. observ.).

Effects of starvation on the flight activity of P. nemorum

To examine whether starvation affects flight activity, 61 virgin males and 62 virgin females of P. nemorum were fed ad libitum for 2-3 d immediately after eclosion. Almost half of them were each separately placed in a test tube ($10 \times 74 \text{ mm}$) only containing water-saturated cotton-wool (male n = 30, female n = 30, "without food"), and the remaining beetles in test tubes with two R. sativus cotyledons and water-saturated cottonwool (male n = 31, female n = 32, "with food"), the control. The test tubes were capped with dry cotton-wool and kept for 5 days at 25 ± 5 °C, 50-70% relative humidity, and a 16L: 8D photoperiod (hereafter called "laboratory conditions") to acclimatize P. nemorum to the new conditions. Then, one P. nemorum individual at a time was placed on a closed Petri dish (70 mm in diameter), which was put in the middle of a semitransparent plastic container [410 (L) × 300 (W) × 220 (H) mm]. The behaviour of the beetle was recorded for 20 min and categorized as either: flying, jumping or walking/resting. When an individual beetle flew or jumped the observation ceased but continued for 20 min when it walked or rested. All observations were conducted between 11:00 AM and 17:00 PM at 25 ± 5 °C. P. nemorum individuals were randomly assigned to a time for observation in order to minimize the effects on their activity. A new Petri dish was used for each observation. To determine whether sex and/or feeding condition had a significant effect on the flight activity of P. nemorum, the number of individuals that jumped and the number of individuals that walked or rested were combined. Then, the data were analyzed using a likelihood-ratio test. When an effect was significant at the 5% level a χ^2 test was used to compare treatments.

Effects of mating status on the activity of female P. nemorum

To determine whether mating status affects female movement 69 virgin adult females and 38 adult males were randomly picked from the stock culture. One female and one male were paired in each of a series of test tubes containing one *R. sativus*

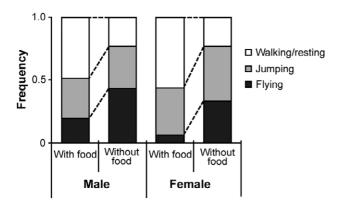


Fig. 1. Effect of starvation on the flight activity of *Phyllotreta nemorum*.

cotyledon and water-saturated cotton-wool (n = 38, "mated female"), and the remaining females were placed singly in a second series of test tubes each containing one R. sativus leaf and water-saturated cotton-wool (n = 31, "virgin female"). These test tubes were kept for 2 d under laboratory conditions. It was not confirmed whether mating occurred during the two days. Then, the P. nemorum females were individually transferred to new test tubes with only water-saturated cotton-wool and kept without food for 5 days under laboratory conditions. The four "mated" females that died during the five days were not included in the analysis. Then, the behaviour of each individual female was observed for 20 min in the same manner as described above. To determine whether female movement is significantly affected by mating status the number of individuals that flew and jumped were combined. Then, the data were analyzed using a χ^2 test.

RESULTS AND DISCUSSION

The flight activity of individuals of *P. nemorum* with and without food differed (P = 0.006, Table 1). Although it did not differ between sexes (P = 0.078), there was a tendency for males to be more flight active than females. In both sexes, more of the individuals that were not fed flew (male 43.3%, female 33.3%; Fig. 1) than of those provided with food (male 19.4%, female 6.3%; male χ^2 = 4.087, P = 0.043; female χ^2 = 7.276, P = 0.007). This indicates that starvation stimulates flight activity in *P. nemorum*. It is likely that hunger stimulates *P. nemorum* to fly in search of food.

The movement of females was affected by their mating status ($\chi^2 = 7.919$, P = 0.005, Fig. 2). The number of mated females that flew or jumped (81.8%) was greater than of virgin females (48.4%). As pupation of *P. nemorum* occurs in the soil, virgin females of *P. nemorum* may stay in areas where conspecific males are more likely to occur. In contrast, mated females do not need to wait for males. Thus, they may move elsewhere

TABLE 1. Results of a likelihood-ratio test of the effect of starvation on the flight activity of *Phyllotreta nemorum*.

	df	χ^2	P
Sex	1	3.108	0.078
Food condition	1	11.878	0.006
Sex \times food condition	1	0.751	0.386

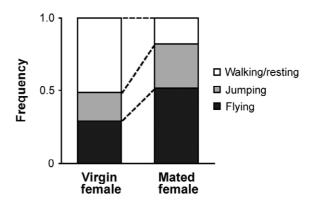


Fig. 2. Effect of mating on the activity of female *Phyllotreta* nemorum.

by flying or jumping. Mated females might need to search for oviposition sites. In the first experiment, virgin males and females were either fed or not fed, while in the second experiment both virgin and mated females were not fed. Therefore, the result for "female without food" in Fig. 1 should be similar to that for "virgin female" in Fig. 2. However, they appear to be different. The females used in these two experiments differed in age. In addition, although the first and the second experiments were done at the same place they were done at different times. These factors may have caused the discrepancy. In any case, the significant difference between treatments in the first experiment is a robust result.

Here, we focused on physiological factors affecting the flight activity of *P. nemorum*. However, the flight activity may be also affected by atmospheric factors, such as wind and temperature (e.g., Israd et al., 1999; Bonsignore & Bellamy, 2007). Moreover, it is suggested that there is gene flow between populations of *P. nemorum* (Nielsen & de Jong, 2005). To better understand the life history of *P. nemorum* it is necessary to determine how long and frequently *P. nemorum* individuals actually fly and/or jump. These points remain to be studied in the future.

ACKNOWLEDGEMENTS. We thank M. Veldkamp and two anonymous reviewers for valuable comments, L. Koopman for rearing plants and T. Mitsunaga for advice on statistics. This study was partly supported by a grant from the Japan Society for the Promotion of Science for Young Scientists (no. 4537) to K.O.

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Received March 23, 2010; revised and accepted April 19, 2010