Effect of Food and Mating on Longevity and Egg Production in the Green June Beetle (Coleoptera: Scarabaeidae)¹

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ABSTRACT A laboratory rearing experiment was conducted to determine the effect of food intake and mating on longevity and egg production of the female adult green June beetle, Cotinis nitida (L.). A positive relationship existed between the number of eggs laid per beetle and beetle longevity (r = 0.37; P < 0.0001), where apple-fed beetles laid 2.2 eggs per day during a lifespan of 23.0 days and the water-fed beetles laid 1.87 eggs per day during a significantly shorter lifespan of 14.8 days (P < 0.0001). After mean adjustments for the effects of beetle longevity (by the LSMEANS procedure), the apple-fed beetles laid an average of 50.8 eggs which was significantly higher than the 26.9 eggs laid by the water-fed beetles (P < 0.0002). The number of matings had no effect on the numbers of eggs produced nor on beetle longevity. The treatment effects on percentage egg hatch by mating and feeding were insignificant. However, the effect of mating on the percentage of egg hatch differed depending on feeding. These differences were presumed to be due to soil differences between egg rearing containers and not to the effects of mating or feeding. These results suggest that feeding beetles live longer and produce more offspring than beetles that do not feed.

KEY WORDS Mating, feeding, longevity, aggregation, *Cotinis nitida*, Coleoptera, Scarabaeidae, egg production, green June beetle.

The green June beetle, *Cotinis nitida* (L.), is native to much of the eastern and south-central U.S. (Goodrich 1966). Pastures and turf areas may harbor large populations of green June beetle larvae that tunnel in the soil and feed on decomposing organic matter. Through this activity, larvae can be economically important in areas such as golf courses and lawns, because they deposit small mounds of soil at the surface openings of their tunnels and may damage plant roots.

From late June to the end of July in Arkansas, adult green June beetle emergence and mating occur in a locally well synchronized manner (J. M. D., unpublished data). Shortly after breaking out of their pupal cells and digging to the surface, female green June beetles release a sex pheromone that attracts numerous males (Domek and Johnson 1987). Mated females return to the soil where they lay eggs, and males continue to fly in search of receptive females. Mating activity persists for 12-14 days at any one location.

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During and after the mating period, beetles may feed on flowers and ripe fruit or scrape the surfaces of tender twigs and herbaceous stems. However, thousands of green June beetles have been observed to emerge annually. They reproduce in pastures covered mainly by dry, senescent grasses where food sources are not sufficient to feed the many beetles present. Despite this apparent limit of food resources, beetles in such areas reproduce successfully, as indicated by the annual emergence of subsequent generations. In contrast, beetles also emerge from pastures located within flying range of plantings of various vegetables and fruits at a time that coincides with ripening. In response to fruit volatiles and aggregation semiochemicals released by feeding green June beetles, additional beetles fly into these areas and form tightly-packed feeding aggregations on fruit. Beetles in a feeding aggregation typically stop feeding, mate and fly off to oviposit in the soil (Domek and Johnson 1988, 1990).

The purpose of this study was to determine if different feeding and mating treatments under laboratory conditions had a significant influence on longevity and egg production of female green June beetles.

Materials and Methods

In May of 1987, about 350 green June beetle pupal cells were collected by digging and sifting pasture soil. Pupal cells were held individually in 8-cm diam. plastic cups (10-cm high) half filled with moist soil and covered with a perforated, clear plastic lid. The cups were kept in the laboratory at 25°C and a photoperiod of 16:8 (L:D). The cups were checked daily for the presence of adult beetles. Beetles were sexed and returned to their respectively labeled containers.

Five 1- to 3-day old females were marked on the pronotum or elytra with a wax pencil and placed separately in 10×8 cm plastic cups. Three unmarked males were placed with each female, and the group was observed until each female had mated. Females were readily mated between 0800 and 1300 hrs (CDT), a time that coincided with flight activity in the field. After mating, one female was placed in each of 100 plastic milk containers (3.8-liter) filled with moist soil to a depth of 10 cm. The females were supplied with distilled water held in a 3-cm high \times 4-cm diam. plastic cup lined with cotton balls. Water was replaced or replenished as needed.

To simulate the abundant food encountered by feeding aggregations of beetles, 50 females placed individually in 50 containers were each given one-fourth of a 'Red Delicious' apple, that was replaced with fresh apple as needed. Individual beetles in the remaining 50 containers received water only, which required females to utilize stored energy sources to sustain reproductive and metabolic activities.

After 7 or 8 days, the soil in all containers was sifted to recover any eggs. The number of eggs was counted and recorded for each female. Eggs were individually placed in labeled 2.5-cm diam. \times 4-cm high glass vials containing moist soil, and were kept at 25°C and a photoperiod of 16:8 (L:D). The numbers of eggs that hatched were noted.

Approximately 9 days after their first mating, 25 of the water-only treated females and 25 of the apple-fed females were mated for a second time, as described above. Females were returned to their respective containers with fresh, moist soil, and water and/or apple slices were replaced as needed. All females were observed daily for activity. After 7 or 8 days, the soil in these containers was sifted to recover any eggs (handled as described above).

Records were made of the total number of eggs laid, the number of eggs that hatched and the lifespan of each female. An analysis of covariance of the 2×2 factorial design was performed to test the significance of treatment effects and to detect possible interactions using the GLM procedure of SAS (SAS Institute 1986). Adjusted means for eggs laid and for longevity were calculated using the LSMEANS statement of GLM (SAS Institute 1986). An analysis of variance for the two-factor factorial experiment was used to analyze the percentage egg hatch.

Results and Discussion

There was positive relationship between the number of eggs laid and beetle longevity (r = 0.37; F = 18.5; df = 1, 95; P < 0.0001). The mean numbers of eggs laid per day were nearly equal for water-fed beetles and apple-fed beetles, i.e., 1.87 and 2.2 eggs laid, respectively (Table 1). However, there was a significant (F = 11.24; df = 3, 95, P = 0.0001) eight day difference between the longevity of apple-fed beetles ((23.0 days) and water-fed beetles (14.8 days). Thus apple-fed beetles laid more eggs than did water-fed beetles because they lived longer. This relationship required that all mean counts of eggs laid be adjusted to remove the effect of longevity.

Treatments*	Mean longevity (days)†	Mean No. eggs laid†	Estimated mean eggs per day‡	Percentage eggs hatched†
Water: 1 mating	$14.9\pm0.9~{ m b}$	$26.7\pm5.2~\mathrm{b}$	1.79	$51.8\pm5.4~\mathrm{b}$
Water: 2 matings	$13.9 \pm 1.0 \text{ b}$	$27.1\pm5.3~\mathrm{b}$	1.94	76.3 ± 5.8 a
Apple: 1 mating	23.1 ± 1.3 a	45.1 ± 5.2 a	1.94	$65.7\pm5.4~\mathrm{ab}$
Apple: 2 matings	23.0 ± 1.3 a	56.5 ± 5.3 a	2.39	57.4 ± 5.2 b

Table 1. A laboratory comparison of the effects of mating frequency and
feeding on female green June beetle mean longevity, number of
eggs laid, eggs laid per day, and percentage egg hatch.

* N = 25 females per treatment.

[†] Column adjusted treatment mean longevity (P 1< 0.002), eggs laid (P < 0.05), and percentage hatch (P < 0.02) followed by a different letter are significantly different as determined by LSMEANS (SAS Institute 1986).

[‡] Treatment mean values were derived from the mean number of eggs laid divided by the mean longevity, but these means were not significantly different.

Apple-fed beetles laid an average of 50.8 eggs which was significantly higher than the 26.9 eggs laid by females with access to water only (F = 14.9; df = 1, 95; P < 0.0002). There was no significant difference (F = 1.62; df = 1, 95; P < 0.23) in the number of eggs laid by females mated once (35.9) and females mated twice (41.8) (Table 1).

Egg hatch was significantly affected (F = 9.11; df = 3, 83; P < 0.003) by the interaction of mating by feeding. Thus as seen in Table 1, the beneficial effect of additional mating when fed apple on the percent of eggs hatched was 8%. In contrast, the beneficial effect for those fed water reached 25%. The effect of limiting food resources, although beneficial for once-mated beetles, reverses itself when beetles are mated twice. These unexplainable effects were perhaps attributed to

unrecorded differences in egg sifting, handling and soil moisture content in the egg rearing containers. The interaction of mating and feeding was not significant (F = 1.24; df = 1, 95; P < 0.27) as far as the number of eggs laid (Table 1).

Similarly, egg production and longevity of female Japanese beetles, *Popillia japonica* Newman (Coleoptera: Scarabaeidae), was significantly influenced by the food plant females consumed. However, continuous mating apparently had no role in maintaining egg fertility (Ladd 1987).

These results suggest that under field conditions individual green June beetles with strictly limited food resources may be constrained to a level of reproduction, longevity and flight activity determined by energy stores carried over from the larval feeding stage. However, individuals that acquire supplemental food, either alone or in feeding aggregations, produce a greater number of eggs, live longer and may have greater dispersal capabilities than individuals with very limited food resources.

An example of this difference in nutritional level may be the extended presence of a green June beetle population near Clarksville, AR which for the past four years was present on fruit until late August. In comparison, a second green June beetle population with very limited resources near Goshen, AR disappeared by mid-July, although these beetles emerge only about a week before those near Clarksville (J. M. D., unpublished data).

Both sexes of green June beetles feed on fruit in aggregations, which also gives the sexes increased access to each other at a time when the primary release of sex pheromone by females and the related major mating activity has passed. Beetles of both sexes engaged in feeding were reported to be significantly more attractive to responding beetles than all other combinations tested (Domek and Johnson 1988). This indicates that the semiochemicals mediating aggregation also organize and direct the post-mating activity of males and females. Individuals that respond to these volatiles to feed/or mate leave more progeny on the average than individuals that do not feed or partake in feeding aggregations. The presence of males in feeding aggregations is presumed to similarly influence their longevity and activity, enabling them to impregnate more females.

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