

## Life History and Demography of *Cephaloleia fenestrata* (Hispinæ: Chrysomelidae: Coleoptera)<sup>1</sup>

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### ABSTRACT

The Neotropical beetle *Cephaloleia fenestrata* is a specialist herbivore on *Pleiotachya pruinosa* (Marantaceae) in upland and flood zone habitat of lowland tropical wet forest in Costa Rica. *Cephaloleia fenestrata* spends its entire life cycle on *P. pruinosa*, feeding primarily in rolled young leaves as adults, feeding in the concavity of leaf petioles as larvae, and laying eggs and pupating on the leaf petioles. Egg development time is about average for Chrysomelidae, approximately ten days. *Cephaloleia fenestrata* larvae, like other rolled leaf hispines, have an extremely long development period (94 d) consisting of only two instars (compared to a range of three to six in other Chrysomelidae). The 30-day pupal stage is also long, but within the range of that observed in other Chrysomelidae. Adults live an average of six weeks. The generation time for *C. fenestrata* is thus approximately six months. Male *C. fenestrata* lived longer and had a higher capture probability than females. There was no difference between males and females in the movement probabilities between upland and flood zone habitat. Rolled *P. pruinosa* leaves are both ephemeral (available to the beetles for approximately two days) and somewhat rare (1 in *ca* 15 ramets has a rolled leaf). Adults must move from leaf to leaf *ca* 36 times during their average adult life span. Still, *C. fenestrata* adults demonstrate high site fidelity.

### RESUMEN

El escarabajo neotropical *Cephaloleia fenestrata* es un herbívoro especialista en *Pleiotachya pruinosa* (Marantaceae) de las partes altas y zonas de inundación de la selva lluviosa tropical de tierras bajas en Costa Rica. *Cephaloleia fenestrata* pasa todo su ciclo de vida dentro de *P. pruinosa*. Como adulto, se alimenta principalmente en el interior de hojas enrolladas jóvenes y como larva en las concavidades de los pecíolos de las hojas. Los huevos y pupas se encuentran en los pecíolos. El tiempo de desarrollo dentro de los huevos es aproximadamente 10 días, cercano al tiempo promedio para la familia Chrysomelidae. Las larvas de *C. fenestra*, como otras de la subfamilia Hispinæ, tienen un periodo de desarrollo exageradamente largo (94 días), consistiendo de solo dos estadios (comparado al rango de tres a seis estadios de otros Chrysomelidae). La etapa pupal de 30 días de duración también es larga, pero está dentro del rango observado en otros Chrysomelidae. Los adultos viven un promedio de seis semanas. Por lo tanto, el tiempo generacional de *C. fenestrata* es aproximadamente seis meses. Los machos de *C. fenestrata* vivieron más tiempo y tuvieron una mayor probabilidad de ser capturados que las hembras. No hubo diferencias entre machos y hembras en cuanto a las probabilidades de movimiento entre las zonas altas y las zonas de inundación. Las hojas enroscadas del *P. pruinosa* son efímeras (están disponibles para los escarabajos aproximadamente durante dos días) y son un tanto escasas (aproximadamente uno de cada 15 tallos tiene una hoja enroscada). Los adultos deben moverse de hoja en hoja aproximadamente 36 veces durante su tiempo de vida promedio. Aun así, los adultos *C. fenestrata* demostraron alta fidelidad de sitios.

*Key words:* *Cephaloleia fenestrata*; *Chrysomelidae*; *Hispinæ*; *Marantaceae*; *mark-recapture*; *Pleiotachya pruinosa*.

THE STRONG ECOLOGICAL RELATIONSHIP OF HISPINE BEETLES (CHRYSOMELIDAE: HISPINÆ) WITH PLANTS IN THE ORDER ZINGIBERALES has a long evolutionary history; their characteristic herbivory damage has been found in the fossil record dating back to the latest Cretaceous (Wilf *et al.* 2000). Known as rolled leaf hispines, these beetles are divided into two tribes: the Cephaloliini and the Arescini. This

diverse group is composed of over 200 described species including 186 Cephaloliini and at least 17 Arescini (Strong 1977a; Staines 1996, 1998); yet, little is known about their demography and the life history of only a handful of species has been studied. We do know that the majority feed, and probably spend their entire life cycle, on Zingiberales. They feed as both larvae and adults by “strip mining” the surface of moist microhabitats formed by various plant structures including primarily the immature rolled leaves (Strong 1977b, Seifert & Seifert 1979), but also detritus-covered mature leaves (McCoy 1984), leaf petioles (Strong 1977a), and

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flower bracts (Seifert & Seifert 1979). Immature leaves of plants in the order Zingiberales emerge rolled in a scroll-like fashion. The rolled leaves provide refuge and food for rolled leaf beetles. Rolled leaves are an ephemeral resource, available to the beetles for only a period of days to a few weeks, depending on the plant species, before they unroll (Strong 1983). In at least one hispine species, adults are long-lived (marked *Chelobasis perplexa* (Arescini) can live for at least 18 mo) and have high site fidelity (Strong 1983). Other species are poorly known. Two species of the Arescini have extremely long larval periods estimated at 201 (Strong & Wang 1977) and 228 days (Seifert & Seifert 1979), although one species of the Cephaloliini has a much shorter larval period of an estimated 32 days (Seifert & Seifert 1979). Rolled leaf beetles live at low population densities; nonetheless, the beetles are easy to locate because the host plants are distinctive and conspicuous elements of the tropical forest understory and the beetles can be found reliably on their hosts. With so few rolled leaf beetle species studied, however, the above generalizations are tentative at best.

The long life span of rolled leaf hispines is puzzling, especially considering that rolled leaves are an ephemeral resource. The average beetle must search for a new rolled leaf many times during its lifetime. Furthermore, only a fraction of plants have rolled leaves at any given time, adding to the difficulty in finding a new rolled leaf. Also, it seems paradoxical that a beetle that must search for a spatiotemporally limited resource would have such a high site fidelity. How the beetles persist in this changing environment is unknown.

The present study is the most complete life history and demographic study of a rolled leaf hispine beetle. In this study, I asked if a specialist rolled leaf beetle, *Cephaloleia fenestrata* Weise, demonstrates life history characteristics similar to other rolled leaf hispines, such as (1) an elongated larval period, (2) a long-lived adult stage, and (3) high site fidelity. I describe the entire life cycle of *C. fenestrata* and discuss the consequences for living in an environment with a patchy ephemeral resource. Moreover, a subset of habitat patches is located in a flood zone, and flooding has been demonstrated to greatly reduce populations of *C. fenestrata* (Johnson in press). I distinguished between population fluctuations associated with seasonality and stochastic flooding events. Also, this is the first study to test for sex-specific differences in life history and spatial demography of a rolled leaf hispine.

To date, I have found *C. fenestrata* as immatures and adults only on the herbaceous monocot *Pleiotachya pruinosa* (Marantaceae: Zingiberales). *Cephaloleia fenestrata* has been reported to feed as adults on *Ischnosiphon* sp. (Staines 1996); however, this is probably the same plant species because of a change in nomenclature for the plant genus *Ischnosiphon* to *Pleiotachya*. *Cephaloleia fenestrata* is an excellent study species for a spatial demographic study because (1) it is a specialist feeder and thus it can be easily located; (2) the adults can be individually marked; (3) population sizes are not overwhelmingly large and a detailed mark-recapture study can be done to describe adult demography; (4) the beetle lives in easily definable habitat, facilitating estimation of habitat-specific demographic parameters; and (5) by raising immatures in the laboratory, I determined that only one other immature rolled leaf beetle, *C. trivitatta* Baly (Staines 1996), is found on *P. pruinosa*, and its eggs and larvae are smaller and more yellow than those of *C. fenestrata*. Thus, I could distinguish *C. fenestrata* immatures.

## METHODS

**STUDY SITE.**—This study was conducted at La Selva Biological Station in Heredia Province, Costa Rica (10°26'N, 83°59'W). La Selva is in a tropical wet forest on the Caribbean slope of the central mountain range at 35 to 137 m elevation (McDade & Hartshorn 1994) and averages ca 4000 mm of annual rainfall (Sanford *et al.* 1994). While La Selva has a dry season from approximately January to April, no month averages less than 100 mm of rainfall; thus, seasonality is mild (McDade & Hartshorn 1994). The field study area was along ca 1.5 km of the Puerto Viejo River, extending up to 1.5 km away from the river. The flood zone extends linearly along the river for the most part and the upland is adjacent to the flood zone. Upland and flood zone patches of *P. pruinosa* are as little as 20 m apart in some locations. Laboratory studies were done in shade houses and an ambient laboratory on the station.

**POPULATION DENSITY AND FLUCTUATION.**—A previous study demonstrated that flooding was a major mortality factor for *C. fenestrata* adults and a controlled experiment suggested the same for larvae (D. Johnson in press). To determine fluctuations in population density in two habitats (upland and flood zone), I surveyed *C. fenestrata* immatures and adults in the concavity of leaf petioles of *P. pruinosa*

on eight occasions at three-month intervals from March 1999 to December 2000. While adults are primarily found in the rolled leaves, they are occasionally found in the concavity of the leaf petioles. The petiole is the stem of the leaf and its concavity runs lengthwise such that a cross section would produce a crescent moon shape. Petioles converge near the base of the plant, similar to banana plants and *Heliconia* spp., each petiole fitting into the concavity of the outward neighboring leaf petiole. Flood events preceded three of the survey periods; the first, fourth, and eighth by three months, one week, three months, respectively. A total of 75 patches were surveyed, 58 in the upland and 17 in the flood zone. A patch was defined as a group of *P. pruinosa* that was greater than 17 m from the nearest conspecifics (a distance selected for a concurrent metapopulation study so that inter-patch migration was 20%). Patches were composed of sub-patches, which were defined as groups of *P. pruinosa* that were at least 5 m from the nearest conspecifics. *Cephaloleia fenestrata* immatures (eggs, larvae, and pupae) are found almost exclusively in the concavity of leaf petioles of mature unrolled leaves. During each sample period, I searched for immatures in the concavity of the leaf petioles of five haphazardly selected ramets in each sub-patch. A ramet is defined as a vertically growing plant that may or may not be a clone of neighboring plants. If the sub-patch consisted of fewer than five ramets, then all of the ramets were examined. The number of each *C. fenestrata* stage was recorded for each ramet.

Densities of each immature stage were defined as the number of individuals per ramet. Means, standard deviations, and coefficients of variance for immature densities were calculated across sampling periods for each immature stage in both the upland and the flood zone habitat. Standard deviations, 95 percent confidence intervals, and significant differences between the flood zone and upland in each sample period were calculated by running 10,000 bootstraps of each data set with replacement.

Adult *C. fenestrata* live primarily in the rolled leaves of *P. pruinosa*. Adults were sampled from *P. pruinosa* rolled leaves every 25 days from January 1999 to January 2000, and then were sampled every three months until December 2000. Data from the first year of this study were used in a mark-recapture study described below. During each sampling period, all rolled leaves with an apex opening of 2 mm or greater were gently opened and the adult beetles were counted and given a unique mark by carefully puncturing the elytra (the hard-

ened forewings of beetles). Puncturing the elytra does not seem to harm the beetles (Unruh & Chauvin 1993; D. Johnson, pers. obs.). Beetles could only be sexed when they were mating. Locations and census periods of captures and recaptures were noted, and the beetles were released into the same rolled leaves from which they were removed.

Sex-specific lengths and widths of adult *C. fenestrata* were measured under a dissecting microscope. I tested for sexual dimorphism in adult size with a *t*-test. Length was measured as the distance from the apex of the head to the posterior of the elytra. Width was the measure of the widest distance across the elytra.

EGG STAGE DURATION AND SURVIVAL.—*Cephaloleia fenestrata* eggs were both marked in the field and collected from the field and brought into the laboratory. Eggs were checked daily or every two days until either they hatched or they were determined to be dead. No egg parasitoids were found. By assuming that eggs were collected at random periods into their development and that reproduction was continuous, I assumed that eggs were on average collected halfway through their developmental period. Thus, I doubled the average time to hatching to estimate the egg development time.

LARVAL GROWTH, STAGE DURATION, AND SURVIVAL.—I measured larval growth rate in the field from May to July 2000, and in a shade house from September 2000 to February 2001. A total of 465 measurements were made of 99 individual larvae. I measured lengths and widths of larvae and the separation of the eyes, every two to five days in the field and every six to ten days in the shade house. Separation of the eyes increases incrementally with changes in instar, and thus was used as an indicator of instar. Size-specific growth rate per day was calculated by dividing the growth, defined as change in length (mm), by the number of days between observations.

Following a larva from hatching to pupation proved difficult because invariably the larva was lost for unknown reasons. So the development time of each instar was estimated indirectly by integrating the reciprocal of size-specific growth rates (mm/d) for the estimated larval sizes over the interval of sizes from the beginning to the end of each instar. The total development time of *C. fenestrata* larvae was then calculated by summing the instar-specific development times. This is a modification of the method used to estimate larval stage duration in

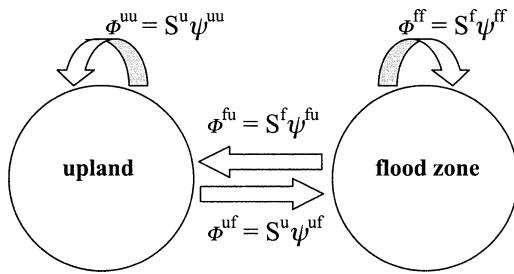


FIGURE 1. Graphical illustration of a two-strata mark-recapture model. The transition probability  $\Phi$  is the product of the survival probability  $S$ , and the movement (or lack of movement) probability  $\psi$  is the probability. Superscripts  $u$  and  $f$  refer to the upland and flood zone, respectively. Thus, the transition probability from the flood zone to the upland is represented as  $\Phi^{uf}$ .

the related rolled leaf hispine *C. perplexa*, which was integrated from hatchling size to pupation size and ignored instar-specific growth rates (Strong & Wang 1977). Larval growth curves were fit using the Curve-fitting Toolbox in Matlab<sup>®</sup>.

**PUPAL SIZE, STAGE DURATION, AND SURVIVAL.**—Pupae were collected both from the field and from the shade house and brought into a laboratory with ambient conditions. Pupal lengths and widths were measured using a dissecting microscope and pupae were checked daily until eclosion, parasitoid emergence or obvious death. By assuming that the collected pupae were a random sample and that larvae pupate continuously over time, I estimated pupal stage duration by doubling the observed mean time to eclosion.

**DEMOGRAPHIC PARAMETERS.**—To examine survival ( $S$ ), capture ( $p$ ), and between-habitat movement ( $\psi$ ) probabilities of *C. fenestrata* adults and to test for sex-dependence in these probabilities, I analyzed mark-recapture data in a multi-strata model design in program MARK version 1.8 (White 2001). Movement was measured between upland and flood zone habitat. In the model,  $\phi$  is the product of survival and movement (or non-movement) probabilities (Fig. 1). For example, the probability of a beetle in the upland ( $u$ ) at time  $t$  being alive in the flood zone ( $f$ ) at time  $t + 1$  is expressed as  $\phi^{uf} = S^u \psi^{uf}$ . The model assumes that the survival of a beetle that moves between strata is the same as that of a beetle remaining in the donor stratum. While I will refer to the parameter  $\psi$  as a movement parameter, whether the beetle moved between strata or stayed in the same strata is in-

dicated by the superscript. For example, the expression  $\psi^{uf}$  indicates the probability that the beetle moves from the upland ( $u$ ) to the flood zone ( $f$ ) over the given time step. Alternatively, the expression  $\psi^{uu}$  indicates the probability that the beetle remains in the upland. Note that all movement probabilities for stratum  $a$ ,  $\psi^a$ , sum to one ( $\sum \psi^a = 1$ ) and all transition probabilities from stratum  $a$  sum to the survival rate in stratum  $a$ , ( $\sum \phi^a = S$ ). Capture probability is defined as the probability of capturing an organism conditional on it being alive and present in that stratum during that sampling period. So the probability of capturing a beetle that was in stratum  $a$  at time  $t$ , in stratum  $a$  at time  $t + 1$  is given by the equation  $P(a_{t+1}|a_t) = P^a(t)S^a(t)\psi^{aa}(t)$ . In this analysis, I used mark-recapture data for those beetles with known sexes. A beetle only needed to be mating during one of its potentially multiple capture events for its sex to be known; thus, measuring differences between male and female demographic parameters was possible. For example, if a male and female were captured while mating at time 1, but then only the male is recaptured at times 2 through 4, and then both are recaptured at time 5, then the male will have a larger capture probability than the female. I tested for dependence of survival, capture, and movement probability on sex and habitat (upland vs. flood zone). Akaike's (1973) information criterion (AIC) was used to select the model that best explained the variation in the data with the fewest parameters. The model with the lowest AIC is the model with the better fit. Herein, I report  $\Delta AIC$  values, which are the differences between the lowest AIC value and the AIC value of a given model. Thus, a  $\Delta AIC = 0$  indicates the best-fit model.

**ROLLED LEAF AVAILABILITY.**—During each census, the number of rolled leaves in the upland and the flood zone was counted. The total number of ramets in each sub-patch was counted in June 1999. The density of rolled leaves was calculated as rolled leaves/ramet.

I classified stages of *P. pruinosa* rolled leaves by two leaf characteristics: the diameter of the opening at the apex ( $<2$  or  $\geq 2$  mm) and how far the opening extended down the leaf (Fig. 2). For example, a leaf in which the apex opening extended down the leaf 2/8 of the way was classified as stage two and one in which it extended 4/8 of the way down the leaf was stage four. Young leaves, in stage zero, could have an apex opening of less than 2 mm (stage 0-a) or 2 mm or greater (stage 0-b). All

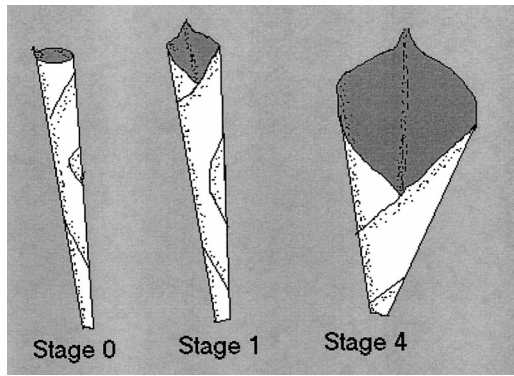


FIGURE 2. Examples of rolled leaf development stages. Stage 0 indicates that the hole at the apex of the leaf extends 0/8 down the leaf, stage 1 indicates that the hole extends 1/8 down the leaf, and so forth.

rolled leaves in stages 1 through 7 had an apex opening of 2 mm or more in diameter.

To determine which rolled leaf stages were utilized by *C. fenestrata*, I destructively sampled rolled leaves in all stages to count the number of adult beetles inside. I determined the amount of time that a rolled leaf was available to the beetles by estimating the amount of time it took a leaf to go through the stages that were used by *C. fenestrata*. Also, I counted the number of rolled leaves in the study area from January 1999 through January 2000. I compared the per ramet density of rolled leaves in the upland to those in the flood zone of the Puerto Viejo River.

## RESULTS AND DISCUSSION

**POPULATION DISTRIBUTION.**—The mean densities of eggs, larvae, and pupae fluctuated throughout the study (Fig. 3). Densities were generally higher in the upland than in the flood zone while the coefficient of variation was higher in the flood zone than in the upland (Table 1). Immature densities were always lower in the flood zone than in the upland in the first sampling period following a

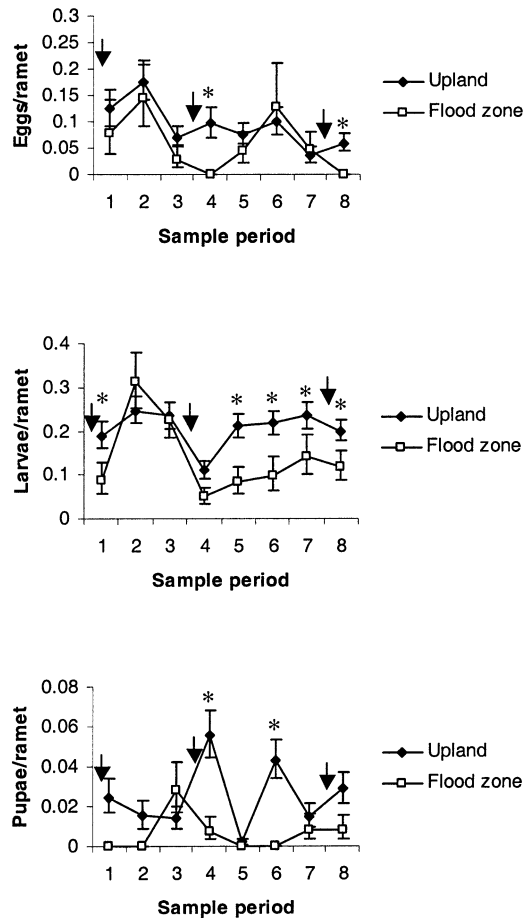


FIGURE 3. Densities (individuals/ramet  $\pm$  1 SE) of *Cephaloleia fenestrata* eggs, larvae, and pupae from March 1999 to December 2000. Arrows indicate that a flood event occurred between the respective sample periods. Asterisks indicate significant differences ( $P < 0.05$ ) between densities in the upland and flood zones during that sample period.

flood (sign test,  $P < 0.05$ ; Ambrose & Ambrose 1987). Egg densities in the flood zone were significantly lower than in the upland in the first sample period following two of the three floods (Fig. 3).

TABLE 1. Densities (individuals/ramet  $\pm$  SD) and coefficients of variation (CV) for the immature stages of *Cephaloleia fenestrata* on *P. pruinosa* in the upland and flood zone habitats.

|        | Upland           |      |   | Flood zone       |      |   |
|--------|------------------|------|---|------------------|------|---|
|        | $\bar{x} \pm$ SD | CV   | N | $\bar{x} \pm$ SD | CV   | N |
| Eggs   | 0.09 $\pm$ 0.04  | 0.47 | 8 | 0.06 $\pm$ 0.05  | 0.92 | 8 |
| Larvae | 0.21 $\pm$ 0.04  | 0.21 | 8 | 0.14 $\pm$ 0.09  | 0.62 | 8 |
| Pupae  | 0.03 $\pm$ 0.02  | 0.70 | 8 | 0.01 $\pm$ 0.010 | 1.50 | 8 |

Larval densities in the flood zone were significantly lower than in the upland in five of the eight sample periods, two of which immediately followed floods (Fig. 3). Pupal densities in the flood zone were significantly lower than in the upland during two of the sample periods, one of which immediately followed a flood (Fig. 3). Thus, in the nine times in which immature densities differed between habitats, densities in the flood zone were always lower. This is consistent with a concurrent experiment that found 100 percent of larvae ( $N = 72$ ) was killed by simulated flooding of 12 hours or more (Johnson in press). The combination of the data from the current study and the simulated flooding experiment strongly indicate that flooding decreases densities of immatures in the flood zone.

Over the two year study, 2755 *C. fenestrata* adults were sampled in 2926 rolled *P. pruinosa* leaves (0.92/rolled leaf, median = 0, range = 0–17). Adults were in the concavities of the leaf petioles at a low density (0.02/ramet). In contrast to my expectation of finding more females than males on the leaf petioles because this was where eggs were laid, I found males and females on the leaf petioles in small but comparable numbers (ten males, seven females,  $\chi^2 = 0.53$ ,  $df = 1$ ,  $P > 0.05$ ). A possible explanation for finding males on the leaf petioles is that they go there to mate with females; however, I reject this hypothesis because none of the 62 *C. fenestrata* found in leaf petioles were mating, a proportion significantly less than the 32.9 percent mating in rolled leaves ( $\chi^2 = 30.11$ ,  $df = 1$ ,  $P < 0.001$ ). An alternate explanation is that adult *C. fenestrata* of both sexes occasionally feed in the concavity of the leaf petioles.

**EGGS.**—The oval yellow eggs were *ca* 2 mm in length and laid in the concavity of the leaf petioles covered with what appeared to be frass. Out of 158 eggs, only 4 percent of *P. pruinosa* ramets ( $N = 3518$ ) contained eggs. Eggs were primarily found singly or in pairs, but were occasionally in groups of up to eight (Fig. 4). Eggs were highly clumped among ramets compared to a Poisson distribution (chi-square goodness of fit test,  $\chi^2 = 618$ ,  $N = 2$ ,  $P < 0.001$ ). It was unclear whether the groups were from a single or multiple ovipositing females. Morrison and Strong (1981) found that the eggs of a congener, which were also significantly clumped, were less likely to be parasitized when in groups; however, I found no evidence of egg parasitoids for *C. fenestrata*. An estimated 23 percent of eggs hatched ( $N = 158$ ) in a field study. The fate of the remaining eggs was unclear. Egg stage

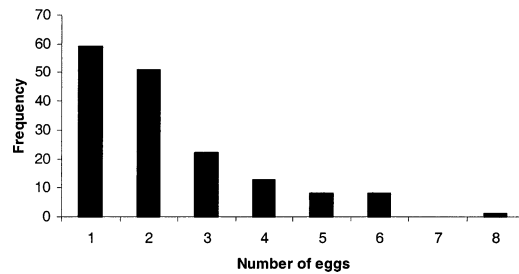


FIGURE 4. Frequency distribution of the number of *Cephaloleia fenestrata* eggs on *Pleiostachya pruinosa* ramets (3379 ramets had no eggs [not shown];  $N = 3518$ ).

duration was an estimated  $10.1 \pm 3.4$  SD days ( $N = 48$ ). A literature review revealed that this egg stage duration was about average for Chrysomelidae ( $\bar{x} = 12.6 \pm 8.3$  SD d, range = 4–35 d,  $N = 17$ ). In a related rolled leaf hispine, *C. perplexa*, egg stage duration was an estimated 19 days (Strong & Wang 1977).

**LARVAE.**—*Cephaloleia fenestrata* larvae are similar to larvae of other rolled leaf beetles: slow-moving, flat, and semitransparent (Strong 1977a). Over the course of the study, I nearly always found *C. fenestrata* larvae feeding in the concavity of the petioles of *P. pruinosa* (862 larvae). In the one exception, a larva was found in a rolled leaf. While I did not raise this particular larva to maturation, its morphology was consistent with that of *C. fenestrata* (and was inconsistent with that of *C. trivittata*, the only other rolled leaf larvae found on *P. pruinosa*). Larvae were 2.66–12.00 mm long and 1.48–6.07 mm wide. The width between the eyes was highly bimodal, strongly suggesting that *C. fenestrata* has two instars (first instar,  $\bar{x} = 0.460$  mm  $\pm 0.013$  SD,  $N = 28$ ; second instar,  $\bar{x} = 0.709$  mm  $\pm 0.017$  SD,  $N = 41$ ;  $t = 63.944$ ,  $P < 0.0001$ ). This was fewer than the range of three to six instars previously reported for Chrysomelidae (Jolivet & Verma 2002). The transition from first to second instar occurred at *ca* 6.22 mm length, and the second instars began at an estimated 7.56 mm. The length/width ratios of first instars differed significantly from second instars (first instar,  $\bar{x} = 1.68 \pm 0.11$  SD,  $N = 28$ ; second instar,  $\bar{x} = 1.98 \pm 0.09$  SD,  $N = 41$ ;  $t = 13.597$ ,  $P < 0.0001$ ).

The growth rate (mm/day) of *C. fenestrata* first instars was a quadratic function of instar length (Fig. 5a;  $N = 381$ ). The growth rate of second instars was a linear function of instar length (Fig. 5b;  $N = 82$ ). First instar: days =  $\int_{3.26}^{6.22} \frac{\text{mm}}{\text{mm}} (0.015x^2$

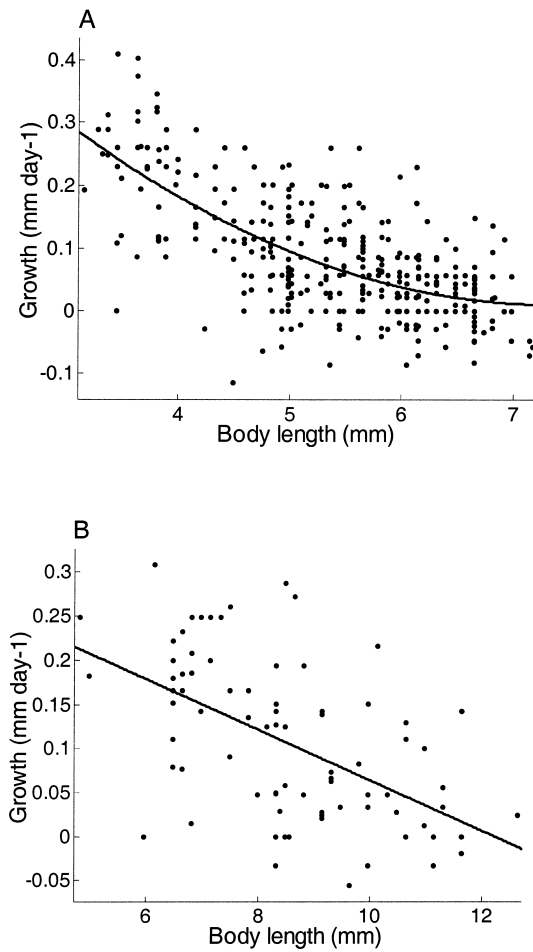


FIGURE 5. Larval growth rates (mm/day) as a function of larval length (mm). (a) Growth rate of first instar larvae is a quadratic function of body length:  $y = 0.016x^2 - 0.22x + 0.771$ . (b) Growth rate of second instar larvae is a linear function of body length:  $y = -0.029x + 0.325$ .

$- 0.224x + 0.8331)^{-1}$  and second instar: days =  $\int_{7.56}^{11.41} \text{mm} (-0.029x + 0.351)^{-1}$ . Larval instar development times were estimated by integrating the reciprocal of the growth rate (d/mm) from estimates of the average minimum and maximum sizes

in each instar (first instar, 3.26–6.22 mm; second instar, 7.56–11.41 mm). The estimated duration of the first instar was 34.52 days, and the estimated duration of the second instar was 60.88 days. Thus, the total larval stage was an estimated 95.40 days, approximately three times longer than that of congener *C. neglecta* (Seifert & Seifert 1979) but less than half the development time of two other rolled leaf hispines in the tribe Arescini (Strong & Wang 1977, Seifert & Seifert 1979). These were unusually long larval development times for Chrysomelidae (see Strong & Wang 1977 for a review).

**PUPAE.**—Pupae look similar to larvae except that their colors are darker (yellowish with maroon), with two series of maroon dots in lateral rows offset from center down each side of the body. All pupae found in the field were in the concavity of leaf petioles. *Cephaloleia fenestrata* pupae were *ca* 10.58 ( $\pm 0.51$  SD) mm long and 5.39 ( $\pm 0.42$  SD) mm wide ( $N = 96$ ). A significant paucity of pupae, as compared to larvae and adults, suggests that some may pupate in locations other than the concavity of the leaf petioles (perhaps in the surrounding leaf litter). Pupal stage duration was an estimated 29.4 days ( $N = 54$ ). A literature review revealed that this pupal stage duration was long ( $>2$  SD away from mean) compared to the durations in other Chrysomelidae ( $\bar{x} = 11.7 \pm 7.9$  SD d, range = 5–35 d,  $N = 15$ ). In a related rolled leaf beetle, *C. perplexa*, pupal duration was an estimated 20 days (Strong & Wang 1977). Pupal survival was not significantly different between the two years ( $\chi^2 = 4.56$ ,  $df = 3$ ,  $P > 0.05$ ); thus, I pooled the pupal survival data. Overall pupal survival probability was 68 percent ( $N = 104$ ). More pupae were parasitized by an unidentified hymenopteran parasitoid from September through November 1999 than from June through September 2000 (Table 2;  $\chi^2 = 117.94$ ,  $df = 3$ ,  $P < 0.001$ ). This suggests that there is temporal variation in parasitoid attack on *C. fenestrata*. It is unclear whether the parasitoids attack the pupae or earlier developmental stages.

TABLE 2. The number of *Cephaloleia fenestrata* pupae parasitized and not parasitized during two sampling periods: September–November 1999 and June–September 2000 ( $\chi^2 = 32.91$ ,  $df = 1$ ,  $P < 0.001$ ).

|                 | <i>C. fenestrata</i> pupae |             |
|-----------------|----------------------------|-------------|
|                 | Not parasitized            | Parasitized |
| Sept.–Nov. 1999 | 8                          | 11          |
| June–Sept. 2000 | 1                          | 84          |

ADULTS.—Adult *C. fenestrata* have an orange–red head and pronotum and black elytra, each with two large yellowish dots. The adults feed primarily on the rolled leaves of *P. pruinosa*, but are secondarily found feeding in the concavity of the leaf petioles. The feeding scars are linear to somewhat curved with well defined to irregular borders. *Cephaloleia fenestrata* prefers the most tender tissue on and near the margin of the lower inner roll of the leaf. In more tender areas, feeding scars completely penetrate the leaf, while in tougher areas the feeding scars are only surface scrapings of the leaf.

Over the course of the year long study, I captured 497 *C. fenestrata* individuals of known sex (249 males and 248 females) a total of 1018 times (the similarity in the number of males and females does not necessarily indicate an equal sex ratio because beetles could only be sexed when they were mating). Female (f) *C. fenestrata* were significantly longer ( $\bar{x}_f = 8.39 \text{ mm} \pm 0.27 \text{ SD}$ ;  $\bar{x}_m = 7.67 \text{ mm} \pm 0.26 \text{ SD}$ ;  $t = 31.01$ ,  $df = 518$ ,  $P < 0.0001$ ) and wider ( $\bar{x}_f = 3.28 \text{ mm} \pm 0.13 \text{ SD}$ ,  $\bar{x}_m = 3.00 \text{ mm} \pm 0.10 \text{ SD}$ ,  $t = 26.59$ ,  $df = 518$ ,  $P < 0.0001$ ) than males (m). There was no difference in the length to width ratio between the sexes ( $\bar{x}_f = 2.56 \pm 0.08 \text{ SD}$ ,  $\bar{x}_m = 2.55 \pm 0.07 \text{ SD}$ ,  $t = 0.84$ ,  $df = 518$ ,  $P < 0.40$ ). Males were captured significantly more times per individual than females ( $\bar{x}_m = 2.38 \pm 1.42 \text{ SD}$ ,  $\bar{x}_f = 1.72 \pm 0.91 \text{ SD}$ ; contingency table,  $\chi^2 = 37.84$ ,  $df = 5$ ,  $P < 0.0001$ ).

DEMOGRAPHIC PARAMETERS.—All demographic analyses were based on a total of 1018 captures of 497 adult *C. fenestrata* ( $N = 249$  males and 248 females). In the most parsimonious model [S(s) p(s,h)  $\psi$ (h)], adult survival probability was sex-dependent (likelihood ratio test,  $\chi^2 = 5.52$ ,  $df = 1$ ,  $P = 0.02$ ), capture probability was sex-dependent ( $\chi^2 = 13.31$ ,  $df = 2$ ,  $P = 0.001$ ) and habitat-dependent ( $\chi^2 = 9.78$ ,  $df = 2$ ,  $P = 0.01$ ), and movement probability was habitat-dependent ( $\chi^2 = 5.88$ ,  $df = 1$ ,  $P = 0.02$ ). Estimates of survival and capture probability are likely overestimates because beetles captured more often were more likely to be sexed (only mating individuals were sexed). Still, sex- and habitat-specific differences in these parameters are unbiased. This model was a better fit to the data than the general model S(s,h) p(s,h)  $\psi$ (s,h), as indicated by  $\Delta\text{AIC} = 5.98$ .

Survival probability was higher in males ( $S_m = 0.79 \pm 0.02 \text{ SE}$  per 25 d) than females ( $S_f = 0.73 \pm 0.02 \text{ SE}$  per 25 d). Capture probability was higher for males than females, and was higher in

the flood zone than in the upland (upland:  $p_m = 0.41 \pm 0.02 \text{ SE}$ ,  $p_f = 0.29 \pm 0.03 \text{ SE}$ ; flood zone:  $p_m = 0.57 \pm 0.06 \text{ SE}$ ,  $p_f = 0.41 \pm 0.07 \text{ SE}$ ). One possible explanation for the lower capture probability of females than males is that females were more often in the concavity of leaf petioles, where they laid eggs. Findings in this study do not support this explanation because similar numbers of males and females were found on the leaf petioles. An alternative explanation is that females were more likely to disperse out of the study area. This explanation would also account for the lower female survival probability. If this were the case, however, I would expect to find that movement probability was greater in females than males. In fact, there was no difference in movement probability between the sexes, suggesting that females did not disperse out of the study area more than males.

Movement probability did not differ between males and females (likelihood ratio test,  $\chi^2 = 2.07$ ,  $df = 2$ ,  $P = 0.36$ ) but was significantly greater from the flood zone to the upland than vice versa ( $\psi^{uf} = 0.016 \pm 0.004 \text{ SE}$ ,  $\psi^{fu} = 0.052 \pm 0.019 \text{ SE}$ ). In a concurrent study, I found that 69 percent of beetles that survived a flood moved from the flood zone to the upland habitat during a flood (D. Johnson in press). Such movement would at least in part explain the difference in movement between the habitats in this study.

ROLLED LEAF AVAILABILITY.—During this study, there was a similar number of rolled leaves per ramet between the upland ( $\bar{x}_{up} = 0.059 \pm 0.012 \text{ SD}$ ,  $N = 17$ ) and the flood zone habitats ( $\bar{x}_z = 0.062 \pm 0.015 \text{ SD}$ ,  $N = 17$ ;  $t$ -test,  $P = 0.48$ ). In the sampling period that immediately followed a flood period, however, plants in the flood zone had 44 percent fewer rolled leaves per ramet than those in the upland (paired  $t$ -test,  $N = 3$ ,  $P = 0.02$ ). Conversely, in periods that did not immediately follow a flood period, there were 23 percent more rolled leaves per ramet in the flood zone than in the upland habitat (paired  $t$ -test,  $N = 14$ ,  $P = 0.01$ ). Thus, the flood zone was initially lacking rolled leaves but later was abundant with rolled leaves compared to the upland habitat.

*Cephaloleia fenestrata* adults utilize rolled *P. pruinosa* leaves with an opening at the apex greater than 2 mm in diameter and with the hole extending 0/8 to 5/8 down the length of the leaf ( $N = 60$ ; Fig. 6). New *P. pruinosa* leaves remain in this rolled leaf condition for only  $ca 1.87 \pm 0.67 \text{ SD}$  days ( $N = 59$ ). Since the average life span of an adult *C. fenestrata* is  $ca 68$  days (based on an esti-



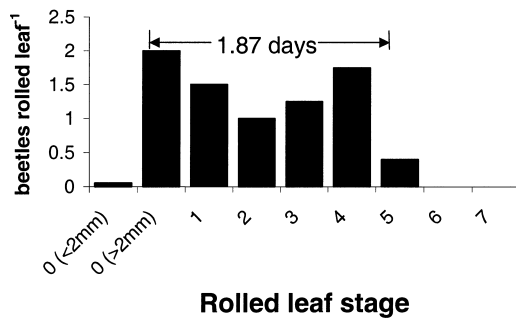


FIGURE 6. *Cephaloleia fenestrata* density during leaves in different stages of rolling. Empirical data indicate that a rolled leaf requires an estimated 1.87 days to progress from stage 0 (>2 mm) to stage 5.

mated survival probability of 0.73 over 25 d,  $N = 1199$ ; D. Johnson in press) and assuming that search time is negligible, I estimate that the average *C. fenestrata* must find a maximum of 36 new rolled leaves during its adult life.

As adults, *C. fenestrata* feed primarily on the rolled leaves, which provide tender tissue and may also provide refuge from predators and food with low toxins. *Cephaloleia fenestrata* adults were found secondarily in the concavity of leaf petioles in a low density. While females oviposit eggs in the leaf petioles, males were found there in similar numbers. Males probably were not going to the leaf petioles to mate because no matings were observed there. Instead, I hypothesize that adults occasionally take refuge in and feed on the petioles.

Development time of eggs is approximately ten days. Upon hatching, larvae feed by strip mining almost exclusively on the leaf petiole, although one larva (probably *C. fenestrata*) was observed in a rolled *P. pruinosa* leaf. The larval period is extremely long (94 d) but consists of only two instars compared to the range of three to six instars reported

for other Chrysomelidae. Strong and Wang (1977) hypothesized that a related rolled leaf beetle had a long larval period because its food source (rolled leaves) was of low nutritional value. Pupal development time is also long for Chrysomelidae, an estimated 29 days, and also occurs in the concavity of the leaf petioles. Additional pupation may also occur in an unknown location. Data herein suggest that parasitoid attack is temporally variable.

*Cephaloleia fenestrata* have an adult life expectancy of an estimated 68 days. In this time, the beetles must track an ephemeral resource, rolled leaves, each of which is only available to the beetles for approximately two days. One out of 15 ramets has a rolled leaf at one time, and the median patch size is only 12 ramets; thus, there is an extreme lack of rolled leaves. Despite the low numbers of rolled leaves per patch, movement between habitats was minimal (1–5%). In another study (Johnson 2003), I looked at movement patterns on a patch to patch basis and found that emigration probability was strongly dependent on patch size; e.g., the probability of dispersing out of a patch with one ramet was estimated at 70 percent per 25 days, compared to a 3 percent probability of dispersing out of the largest patch size of 735 ramets. I hypothesize that *C. fenestrata*'s inability to find a rolled leaf within a patch will lead it to (1) disperse to another patch or (2) retreat to the concavity of a leaf petiole. The leaf petiole may provide a good refuge when rolled leaves are unavailable.

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