Adaptive Significance of Sibling Egg Cannibalism in Coccinellidae: Comparative Evidence from Three Species

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ABSTRACT We examined the nature of sibling egg cannibalism behavior and its developmental consequences in three aphidophagous Coccinellidae that all lay clustered eggs: *Cycloneda sanguinea* L., *Harmonia axyridis* (Pallas), and *Olla v-nigrum* Mulsant. There was variation among species in the proportion of eggs laid singly versus in clusters (*C. sanguinea* > *H. axyridis* ≈ *O. v-nigrum*), the synchrony of egg hatch within clusters (*H. axyridis* > *C. sanguinea* ≈ *O. v-nigrum*), and the proportion of eggs cannibalized by early-hatching larvae (*O. v-nigrum* > *C. sanguinea* > *H. axyridis*). Single eggs were cannibalized at lower rates than clustered eggs in all three species, and cannibals delayed dispersal from clusters until all unhatched eggs were consumed. In all three species, the number of eggs cannibalized was negatively correlated with the proportion hatching synchronously (within 10 min). Egg cannibals survived to adulthood at the same rate as noncannibal larvae in *H. axyridis* and *O. v-nigrum*, and at a higher rate in *C. sanguinea.* In all three species, larvae that cannibalized eggs as neonates molted to the second instar sooner than did their noncannibalizing counterparts, regardless of gender. This translated into reduced total developmental time for both sexes in *H. axyridis*, but only for males in *C. sanguinea* and only for females in *O. v-nigrum.* Adult females weighed significantly more than adult males in all three species and female cannibals were heavier as adults than were noncannibalizing females in *H. axyridis* and *O. v-nigrum*, whereas males had similar weights. Egg cannibalism had no effect on adult weight in *C. sanguinea*, but the rearing diet was not optimal for this species. We conclude that the benefits of sibling egg cannibalism accrue in a gender-specific manner such that females potentially gain more than males, but only if the subsequent larval diet is of sufficiently high quality.

KEY WORDS cannibalism, egg clusters, *Cycloneda sanguinea*, *Harmonia axyridis*, *Olla v-nigrum*

CANNIBALISM IN INSECTS IS a diverse behavior but most often takes the form of predation on vulnerable, conspecific life stages such as eggs, larvae, or pupae by active, larger, or more developed life stages (Elgar and Crespi 1992). Sibling cannibalism is a special case thought to be generally less adaptive because of the loss of inclusive fitness associated with killing a relative. Kin recognition systems may evolve in cannibalistic species, including coccinellids (Joseph et al. 1999), and function to reduce the probability of sibling cannibalism. However, the infertile "trophic eggs" produced specifically as packages of food by worker ants of many species (Hölldobler and Wilson 1990) represent a highly evolved form of cannibalism that is in no way reduced by the relatedness of cannibal and victim. The egg-clustering behavior of female coccinellids may represent a maternal strategy to facilitate sibling egg cannibalism among progeny and thereby improve the survival of early-hatching offspring that must disperse and search for suitable prey (Dixon 2000).

Sibling cannibalism is perhaps best documented in predatory birds and is thought to represent a mechanism for improving offspring survival under conditions of food limitation (Polis 1981). Siblicide seems to be obligate in at least 28 species of avian raptors (Watson et al. 1999). In these species, hatching is typically asynchronous with the second hatching offspring serving as a parental insurance policy (Anderson 1990) that is invariably killed and eaten by the first hatching chick, unless the latter should fail to survive. In other bird species, siblicide may be facultative, and the younger nestling cannibalized only if the food supply is inadequate for both to survive (Estes et al. 1999). This has become known as the "icebox hypothesis" (Alexander 1974) and has been proposed to apply also to cannibalism in coccinellids (Dixon 2000).

Sibling cannibalism in birds is thought to be facilitated by asynchronous hatching of eggs (Amundsen and Stokland 1988, Bryant and Tatner1990), and the same may also be true for lady beetles. However, the situation for coccinellids is not complicated by the partitioning of parental investment among offspring as it is in birds. In the absence of any parental care, the only food a coccinellid mother can provide her offspring is in the form of infertile or late-hatching eggs.

Therefore, female coccinellids could potentially influence opportunities for sibling egg cannibalism among their progeny, not only by laying eggs in clusters but also by laying a portion of infertile eggs, or by somehow manipulating hatching synchrony within clusters.

There are reasons to expect the adaptive significance of egg cannibalism in the Coccinellidae to be very different from that of larval cannibalism. For example, Michaud (2003) demonstrated that the nutritional benefits of egg cannibalism are greater than those of larval cannibalism by rearing larvae of three coccinellid species exclusively on conspecific life stages. In two cases, a diet of conspecific larvae significantly reduced survival, and in all three cases developmental time was increased and adult weight reduced relative to a control diet (eggs of the flour moth, *Ephestia kuehniella* Zeller, supplemented with bee pollen). In contrast, a diet of conspecific eggs actually improved survival and developmental time in one species, *Cycloneda sanguinea* L., with no effects on developmental time and only marginal reductions in adult weight resulting in the other two. This difference likely arises because the protein contained in eggs is in a less differentiated state than that in larvae and is therefore more easily and efficiently assimilated. Larvae are also capable of defensive or evasive behavior, and this increases the risk and/or cost of larval cannibalism relative to egg cannibalism. On these grounds, it seems reasonable to expect that the net benefits of egg cannibalism will usually be higher than the net benefits of larval cannibalism.

Given that an egg cluster is an attractive food item for both adult and larval coccinellids, egg-clustering species typically oviposit outside prey aggregations such as aphid colonies (Kawai 1978) and avoid those that contain conspecific larvae (Dixon 2000). Osawa (1989) showed that the probability of cannibalism increased for egg clusters of *Harmonia axyridis* (Pallas) in proportion to their proximity to prey aggregations. So when should a female facilitate sibling egg cannibalism among her progeny? The answer depends on how egg cannibalism among progeny affects the maternal fitness function. The apparent trade-off is between offspring quality that may be improved by egg cannibalism and offspring number that may be directly reduced by egg cannibalism. For example, Osawa (1992) showed that the maternal payoff for sibling egg cannibalism in *H. axyridis* varies with local prey density and is higher under conditions of low prey availability, as would be expected. Thus, the clustering of eggs represents an insurance policy for females buying uncertain futures for their progeny: it raises minimum maternal fitness under scenarios of limited food availability at the expense of maximum maternal fitness that might be achieved if all offspring survived.

In addition to laying clustered eggs, there are other ways female coccinellids might influence rates of sibling egg cannibalism. They could produce a fraction of infertile eggs to serve as food, or skew the developmental rate of eggs within clusters to generate hatching asynchrony. Alternatively, a proportion of eggs might be protected to some degree from sibling cannibalism by laying them singly. We formulated a series of hypotheses based on our inferences of the adaptive significance of egg cannibalism behavior and its potential benefits. We hypothesized 1) that eggs laid singly would be less likely to suffer cannibalism by siblings than those laid in clusters. We also hypothesized that neonate larvae would 2) exhibit delayed dispersal from clusters whenever late-hatching or infertile eggs were available for cannibalism, 3) consume all unhatched eggs before dispersing, and 4) gain measurable developmental benefits from egg cannibalism compared with siblings denied similar opportunities. Furthermore, we hypothesized that 5) females, being more resource-sensitive than males, would benefit disproportionately from egg cannibalism behavior.

We designed two experiments to test these hypotheses using three coccinellid species that all lay clustered eggs: *H. axyridis*, *C. sanguinea*, and *Olla v-nigrum* Mulsant. Both *C. sanguinea* and *O. v-nigrum* are species indigenous to North America, whereas *H. axyridis* is an introduced species of Asian origin (Michaud 2002). All three are currently sympatric within Florida citrus groves where they share many pests as prey such as *Toxoptera citricida* (Kirkaldy), *Aphis spiraecola* Patch, and *Diaphorina citri* Kuwayama, among others. The three coccinellid species vary considerably in their ability to use these prey for development and reproduction (Michaud 2000, Michaud and Olsen 2004) but have similar seasonality in citrus groves, reproducing mostly in spring and fall when new citrus growth favors population increase of their prey. Cannibalism by larvae, whether of eggs or other larvae, seems to be a common behavior of these species and may have important consequences for life history traits that influence adult fitness.

Materials and Methods

Insects. Stock Colonies of *C. sanguinea*, *H. axyridis*, and *O. v-nigrum* were established from adults that were field-collected in citrus groves in Polk County, Florida, in March 2002. Ovipositing females used in experiments ($n = 30 \pm 5$ per species) were never more than five generations removed from their field-collected ancestors. Both stock colonies and experiments were held on a laboratory bench at a constant temperature of 24 ± 2°C under fluorescent lighting set to a photoperiod 16:8 (L:D) h. Adult beetles were maintained in 1 liter, wide-mouth mason jars filled with shredded wax paper and covered with muslin. Beetles in jars were fed a combination of bee pollen and frozen eggs of *E. kuehniella*, with water continuously available on a cotton wick. Ovipositing females were removed from jars, isolated in plastic petri dishes (5.5 cm in diameter by 1.0 cm) and provided with frozen *Ephestia* eggs and water encapsulated in polymer beads or "hygrocapsules" (Analytical Research Systems, Gainesville, FL) as required. Eggs, typically glued to the inner surface of the petri dish, were collected daily and placed in a Plexiglas incubator until eclosion 3–4 d later. Larvae were reared in petri dishes (as described above) on eggs of *E. kuehniella.*

Experiment 1. Because females distribute their eggs in various spatial patterns in petri dishes, we were forced to distinguish between egg "clusters" and egg "clutches." The daily reproduction (=clutch) of each ovipositing female ($n = 40$–50 for each species) was collected daily by simply removing the female to a clean dish, leaving her eggs adhering to the plastic. The eggs within each dish were counted, their distribution (as singles or in clusters of varying sizes) recorded, and they were then held in the incubator until eclosion 4–5 d later. A cluster was defined as any contiguous group of two or more eggs and a "single" as any egg laid without contacting an adjacent egg. One limitation of conducting such experiments in petri dishes relates to a constraint on spatial scale: single eggs are necessarily laid within 5 cm of clusters, whereas in a field situation they might well be placed at greater distances. However, the forced proximity of single eggs to clusters means that differences in their fate observed in this experiment are, in fact, conservative estimates of differences likely experienced in nature.

Beginning on the fourth day, eggs were checked regularly for evidence of melanization, an indication that hatching was imminent, and then every 15–20 min thereafter until hatching began. Upon eclosion of the first larva in a dish, the time was recorded and a stopwatch was started. Because in all three species a majority of fertile eggs within a clutch tend to hatch synchronously, all larvae eclosing within 10 min of the first hatch were tallied as "synchronously hatching" larvae and those hatching subsequently as "late hatching." On the basis of periodic observations (maximum interval was 1 h during daylight hours), we estimated the total time required for neonate larval dispersal from egg clusters in each clutch as the period from first hatch to the time the last hatching larva abandoned its cluster to explore the petri dish. The number of eggs cannibalized within each clutch was recorded at this time. For each species, larval dispersal times were compared between clutches with and without egg cannibalism using a one-way analysis of variance (ANOVA) (StatSoft 2000).

A test of proportions (StatSoft 2000) was used to compare proportions of total eggs laid as singles versus in clusters among the three species, proportions of clutches cannibalized, and proportions of single versus clustered eggs cannibalized. We also analyzed the relationship between clutch size and the proportion of eggs laid singly for each species by nonlinear regression (SPSS Inc. 1998), by using data only from replicates in which single eggs occurred. We used linear regression (SPSS Inc., 1998) to analyze relationships between numbers of eggs cannibalized and the proportion of clutches that hatched synchronously, and the proportion of eggs cannibalized in clutches of different sizes.

Experiment 2. In this experiment, we monitored hatching egg clusters of the three species and separated larvae into two groups. Noncannibal, "control" larvae were isolated in individual petri dishes (as described above) immediately upon eclosion, as soon as melanization was complete, but before the larva had any opportunity for egg cannibalism. Larvae observed feeding on one or more conspecific eggs shortly after eclosion were designated as "cannibals" and were left in their dishes to complete this behavior undisturbed before they were isolated as described above. A number of cannibals consumed more than one conspecific egg, but this was not quantifiable because clusters often contained several cannibals feeding simultaneously. Both groups of larvae were then reared out to pupation on an ad libitum diet of frozen *Ephestia* eggs with water continuously available encapsulated in polymer beads. Developing larvae were observed every 1–1.5 h between 6:00 a.m. and 10:00 p.m. to record molting times as accurately as possible. Larvae that molted overnight were assigned the 6:00 a.m. time. Exuviae were removed immediately after every molt. Mortality data were compared between control and cannibal larvae with a chi-square goodness-of-fit test. Total developmental time was tallied in hours for each insect as the time from eclosion of the egg to emergence of an adult. Upon emergence, adults were transferred to labeled glass vials and frozen. Subsequently,

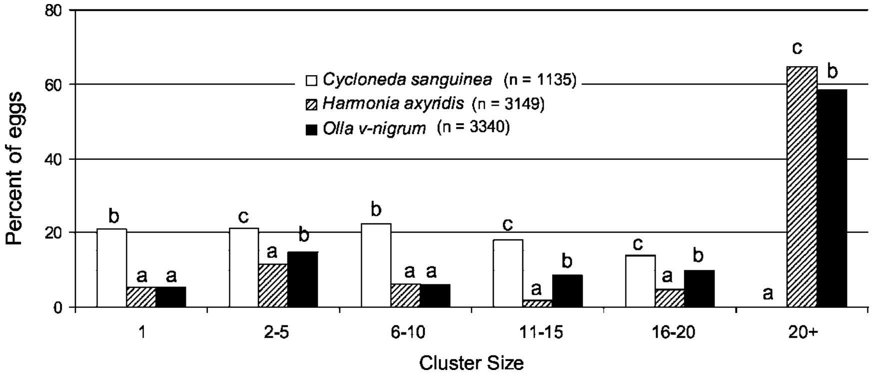

Fig. 1. Proportions of total eggs laid in clusters of different sizes by females of three coccinellid species. Bars bearing the same letter were not significantly different among species for a given cluster size (test of proportions, $P < 0.001$).

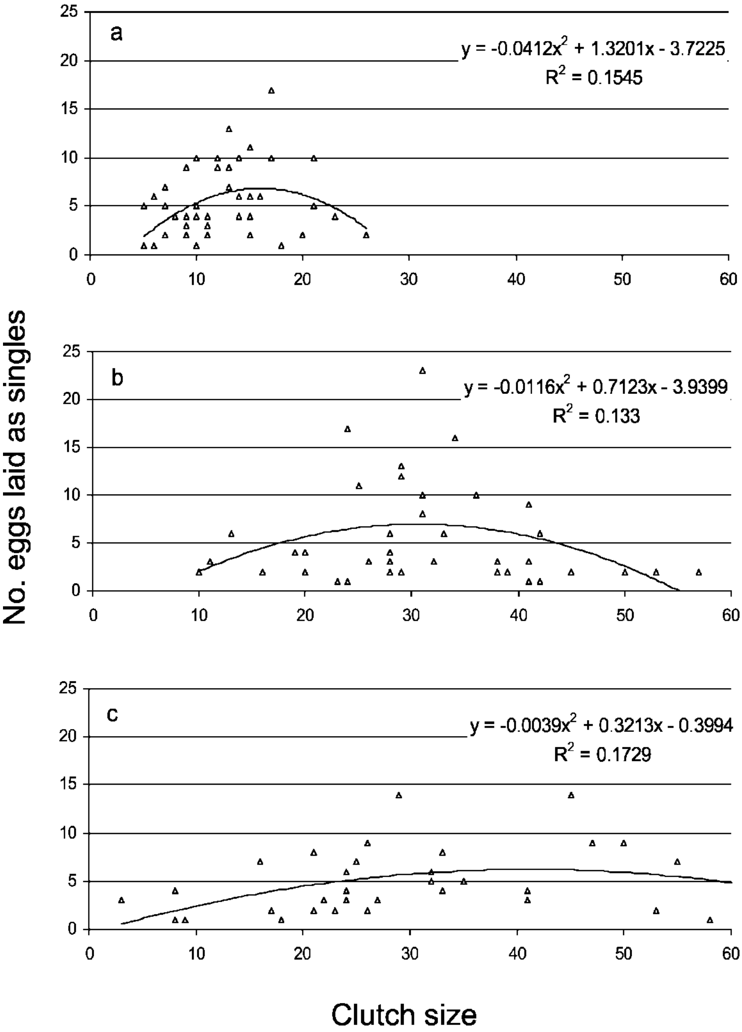

Fig. 2. Numbers of eggs laid singly by females of three coccinellid species plotted against clutch size for replicates in which single eggs occurred. (a) *C. sanguinea.* (b) *H. axyridis.* (c) *O. v-nigrum.* Nonlinear regressions were significant in all cases ($F = 3.34$; df = 1, 41; $P = 0.045$; $F = 4.13$; df = 1, 36; $P = 0.050$; and $F = 3.66$; df = 1, 35 df; $P = 0.036$, respectively). Significance levels for second order coefficients were $P = 0.1131$, 0.0132, and 0.0360, respectively.

adults were defrosted and dissected to verify sex and then dried in a 50°C oven for 3 d before weighing on an analytical balance. A chi-square goodness-of-fit test was used to check sex ratios for deviation from an expected value of 1:1. Data for first instar duration, total developmental time and adult weight were analyzed using a two-way ANOVA (StatSoft 2000) with gender and treatment as independent variables. This was followed by single factor ANOVAs of developmental time and adult weight by gender, and by treatment for each gender. Differences with $P > 0.05$ were assumed to be not significant.

Results

Experiment 1. Females of *H. axyridis* and *O. v-nigrum* laid more than one-half of their eggs in clusters of 20 or more, whereas *C. sanguinea* females produced no clusters this large, but they laid a significantly larger proportion of their eggs as singles (Fig. 1). The relationship between clutch size and the numbers of eggs laid as singles was parabolic in all three species (Fig. 2); a negative quadratic equation provided a significant ($P < 0.05$) nonlinear description of the data for all three species, and second order coefficients were also significant for *H. axyridis* and *O. v-nigrum,* but not for *C. sanguinea.* A total of 137 of 1,135 *C. sanguinea* eggs seemed infertile and neither hatched nor were consumed by siblings. Comparative numbers were 140 of 3,149 eggs for *H. axyridis* and 190 of 3,430 eggs for *O. v-nigrum.* In most cases, these infertile eggs desiccated completely before any hatching of viable eggs. Because these eggs comprised a portion of the total reproduction, but they were neither viable nor suitable as food for cannibalism, they were included in the number of total eggs comprising clutch size, but they

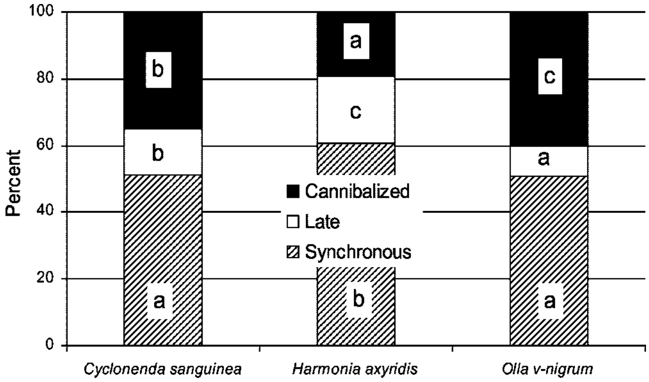

Fig. 3. Percentages of fertile eggs of three coccinellid species that hatched synchronously (within 10 min of first hatch), late (>10 min after first hatch), or were cannibalized by siblings. Bars bearing the same letter were not significantly different among species within a category (test of proportions, $P > 0.05$).

were omitted from representation in Fig. 3, which depicts only the fate of potentially fertile eggs.

In all three species, the probability of cannibalism was approximately twice as high for clustered eggs as it was for single eggs (*C. sanguinea,* 30.6 versus 16.9%; *H. axyridis,* 18.4 versus 7.5%; *O. v-nigrum,* 34.6 versus 14.9%; $P < 0.001$ in all cases). Sibling egg cannibalism was highest in *O. v-nigrum* and lowest in *H. axyridis,* with *C. sanguinea* intermediate ($P < 0.01$ in all cases; Fig. 3). A larger proportion of late-hatching eggs survived to eclose in *H. axyridis* clusters than in *C. sanguinea* clusters, and the latter, in turn, had more late-hatching survivors than did *O. v-nigrum* clusters ($P < 0.001$ in all cases). A larger proportion of eggs hatched synchronously in *H. axyridis* clutches than in clutches of *C. sanguinea* or *O. v-nigrum* ($P < 0.001$ in both cases), with no difference between the latter two species. In all three species, the number of eggs cannibalized displayed a significant negative correlation with the proportion of eggs hatching synchronously in a clutch (*C. sanguinea:* $F = 20.83$; df = 1, 91; $P < 0.001$; *H. axyridis:* $F = 53.654$; df = 1, 96; $P < 0.001$; *O. v-nigrum:* $F = 36.590$; df = 1, 114; $P < 0.001$; Fig. 4).

Although sibling egg cannibalism was common in clutches of all three species, it varied in frequency. In *O. v-nigrum,* 94.1% of clutches exhibited sibling egg cannibalism, significantly more than the 78.7% of *H. axyridis* clutches (test of proportions, $P < 0.001$). Cannibalism occurred in 87.5% of *C. sanguinea* clutches, not significantly different from either of the other species ($P > 0.05$). In all three species, complete dispersal of larvae within clutches was delayed by ≈5 h when eggs were available for cannibalism compared with when they were not (*C. sanguinea:* 28.5 ± 0.4 versus 23.6 ± 1.0 h, $F = 20.663$; df = 1, 86; $P < 0.001$; *H. axyridis:* 24.0 ± 0.5 versus 18.9 ± 0.6 h; $F = 20.326$; df = 1, 90; $P < 0.001$; *O. v-nigrum:* 26.0 ± 0.3 versus 20.7 ± 0.6 h; $F = 17.325$; df = 1, 150; $P < 0.001$). Linear regressions of proportion of eggs cannibalized on clutch size were not significant for any species (*C. sanguinea:* $F = 0.107$; df = 1, 91; NS; *H. axyridis:* $F = 0.187$; df = 1, 96; NS; *O. v-nigrum:* $F = 1.089$; df = 1, 114; NS).

Experiment 2. Cannibalizing larvae survived to adulthood at a rate similar to noncannibalizing larvae (chi-square, NS) in both *H. axyridis* (94.1 versus 93.6%) and *O. v-nigrum* (94.5 versus 97.2%). However, in *C. sanguinea,* more cannibalizing larvae emerged as adults than did noncannibalizing larvae (93.7 versus 85.7%; chi-square = 5.146, $P < 0.05$). Egg cannibalism shortened the first instar for males and females of all three species, whereas total developmental time was shortened for male *C. sanguinea,* female *O. v-nigrum,* and for both sexes in *H. axyridis* (Fig. 5). Female cannibals of both *H. axyridis* and *O. v-nigrum* were heavier as adults than were noncannibals, but cannibalism had no effect on adult weight in *C. sanguinea* (Table 1). Only the *O. v-nigrum* cohort had a sex ratio that deviated significantly from 1:1 in favor of females (0.73; chi-square = 14.841, $P < 0.001$).

C. sanguinea. The two-way ANOVA revealed that the egg cannibalism treatment significantly reduced the duration of the first instar ($F = 48.728$; df = 1, 109; $P < 0.001$) independent of gender ($F = 0.877$; df = 1, 109; NS), and without any gender × cannibalism interaction ($F = 1.054$; df = 1, 109; NS). There was a significant gender × cannibalism interaction for total developmental time ($F = 4.618$; df = 1, 109; $P = 0.034$). Gender had a significant effect on adult dry weight ($F = 8.021$; df = 1, 109; $P = 0.006$), but cannibalism did not ($F = 0.077$; df = 1, 109; NS), and the gender × cannibalism interaction was not significant for adult weight ($F = 0.723$; df = 1, 109; NS).

The single factor analysis revealed that *C. sanguinea* females took longer to develop than did males ($F = 6.018$; df = 1, 111; $P = 0.016$) and were significantly heavier as adults ($F = 7.889$; df = 1, 111; $P = 0.006$). Cannibalism shortened total developmental time by 5.8% in males ($F = 14.118$; df = 1, 49; $P < 0.001$; Fig. 4), but it had no effect on their weight as adults (Table 1). Cannibal females were not significantly different from noncannibal females in total developmental time ($F = 0.574$; df = 1, 60; NS; Fig. 4) or adult weight (Table 1).

H. axyridis. Once again, egg cannibalism significantly reduced the duration of the first instar ($F = 145.346$; df = 1, 148; $P < 0.001$), although there was also

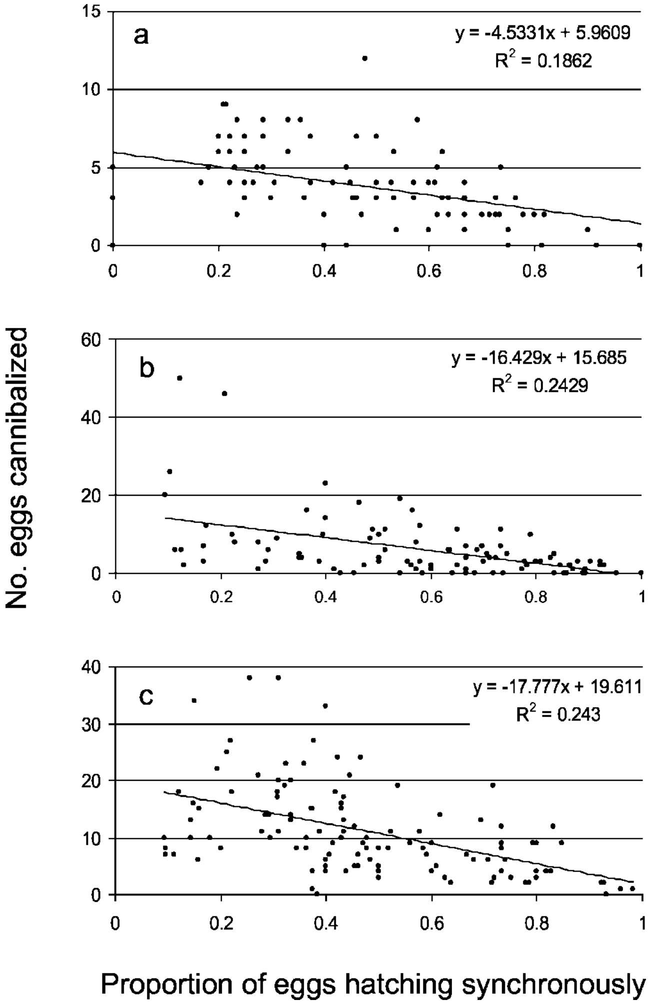

Fig. 4. Proportions of eggs cannibalized plotted against clutch size for females of three coccinellid species. (a) *C. sanguinea*. (b) *H. axyridis*. (c) *O. v-nigrum*. Regression lines were significant ($P < 0.001$) in all cases.

a significant effect of gender ($F = 11.541$; df = 1, 148; $P < 0.001$), and a significant gender × cannibalism interaction ($F = 8.035$; df = 1, 148; $P = 0.005$). Cannibalism significantly reduced total developmental time ($F = 56.004$; df = 1, 148; $P < 0.001$) without an effect of gender ($F = 1.167$; df = 1, 148; NS) or a significant gender × cannibalism interaction ($F = 0.045$; df = 1, 148; NS). Cannibalism significantly increased adult dry weight ($F = 7.950$; df = 1, 148; $P = 0.005$) as did female gender ($F = 14.346$; df = 1, 148; $P < 0.001$), but without a significant interaction between the two factors ($F = 0.355$; df = 1, 148; NS).

The single-factor analysis revealed that females did not develop any faster than males ($F = 1.103$; df = 1, 152; NS) but were significantly heavier as adults ($F = 12.663$; df = 1, 150; $P < 0.001$). Cannibalism reduced total developmental time by 2.5% in males ($F = 33.123$; df = 1, 66; $P < 0.001$) and 4.2% in females ($F = 25.435$; df = 1, 82; $P < 0.001$) (Fig. 4). Female cannibals weighed 6.9% more as adults than did noncannibal females, whereas the 4.9% increase for males was not significant (Table 1).

O. v-nigrum. Egg cannibalism again significantly reduced the duration of the first molt ($F = 34.232$; df = 1, 135; $P < 0.001$) without an effect of gender ($F = 0.876$; df = 1, 135; NS) or a significant gender × cannibalism interaction ($F = 1.000$; df = 1, 135; NS). Total developmental time was reduced by cannibalism ($F = 20.258$; df = 1, 135; $P < 0.001$) without an effect of gender ($F = 0.059$; df = 1, 135; NS) or a significant gender times] cannibalism interaction ($F = 0.439$; df = 1, 135; NS).

In the single-factor analysis, females were significantly heavier than males ($F = 25.877$; df = 1, 136; $P < 0.001$), but they did not differ in developmental time ($F = 0.488$; df = 1, 137; NS). Cannibal males developed 5.0% faster than noncannibal males and weighed an average of 8.5% more as adults, but neither difference was significant ($F = 2.345$; df = 1, 35; NS and $F = 1.592$; df = 1, 35; NS, respectively). However, cannibal fe-

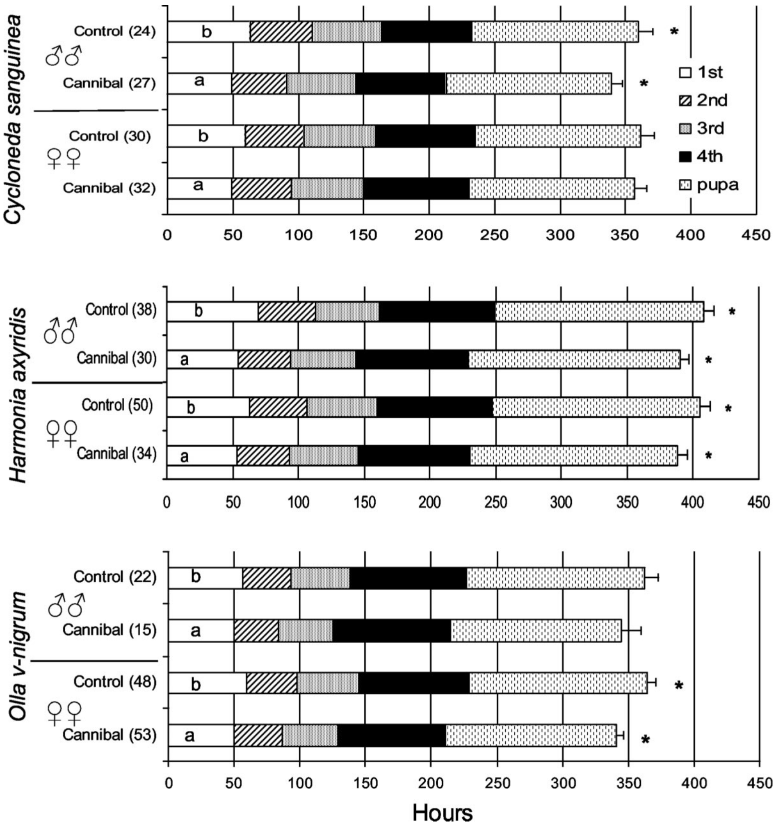

Fig. 5. Mean developmental times (+ cumulative SEM) for immature stages of male and female cannibals and noncannibals of three coccinellid species. Different letters indicate differences in duration of the first instar between cannibals and noncannibals for a particular species and gender (ANOVA, $P < 0.05$). Asterisks indicate significant differences in total developmental time between cannibals and noncannibals within a species and gender. Sample sizes are given in parentheses.

males developed 6.6% faster than control females ($F = 42.228$; df = 1, 100; $P < 0.001$) and weighed 11.1% more as adults ($F = 12.098$; df = 1, 99; $P < 0.001$).

Regressional Analyses. Total developmental time displayed a significant negative correlation with adult weight in both sexes of all three species (*C. sanguinea* males: $F = 13.374$; df = 1, 49; $P < 0.001$; $r^2 = 0.214$; *C. sanguinea* females: $F = 16.612$; df = 1, 60; $P < 0.001$; $r^2 = 0.214$; *H. axyridis* males: $F = 13.989$; df = 1, 66; $P < 0.001$; $r^2 = 0.175$; *H. axyridis* females: $F = 8.110$; df = 1, 82; $P = 0.006$; $r^2 = 0.090$; *O. v-nigrum* males: $F = 40.347$; df = 1, 35; $P < 0.001$; $r^2 = 0.535$; *O. v-nigrum* females: $F = 8.056$; df = 1, 99; $P = 0.006$; $r^2 = 0.075$).

Discussion

We hypothesized that 1) single eggs would be less vulnerable to cannibalism by siblings than clustered eggs, 2) larvae would delay dispersal when eggs were available for cannibalism, and 3) all unhatched eggs would be consumed before complete dispersal of larvae from clusters. All three hypotheses were supported by the results of the first experiment. Single eggs were about one-half as likely to be cannibalized as clustered eggs, even though they were within 5 cm of hatching clusters. Females in nature are not so confined and are likely to distribute single eggs over

Table 1. Mean adult dry weights (±SEM) of male and female cannibals and noncannibals of three coccinellid species

Species	Gender	Adult dry weight (mg)		F	df	P
		Cannibal	Noncannibal			
C. sanguinea	Male	34.3 ± 1.26	33.1 ± 1.26	0.490	1,49	n.s.
	Female	36.5 ± 0.97	37.2 ± 0.95	0.213	1,60	n.s.
H. axyridis	Male	64.1 ± 1.67	61.0 ± 1.79	3.337	1,66	n.s.
	Female	70.4 ± 1.89	65.5 ± 2.20	5.109	1,82	0.026
O. v-nigrum	Male	38.7 ± 2.09	35.7 ± 1.34	1.592	1,35	n.s.
	Female	46.0 ± 1.01	41.4 ± 0.82	12.098	1,99	<0.001

Cannibals consumed one or more conspecific eggs in the first instar, whereas noncannibals did not.

greater distances, even further removed from sibling clusters. Schellhorn and Andow (1999) presented field data to suggest that species-specific patterns of oviposition behavior influence the intensity of egg cannibalism in *Adalia bipunctata* L. and *Coleomegilla maculata* DeGeer as a function of location on the plant and proximity to prey aggregations (aphids).

Complete dispersal of larvae from clusters with eggs available for cannibalism took ≈5 h longer than from clusters without such eggs because cannibals remained until all unhatched eggs were consumed, often becoming torpid and visibly engorged with yolk because a significant proportion of eggs in clusters (20–40%) seem destined to serve as food for early-hatching siblings, this implies potentially significant costs to maternal fitness. However, these costs may not be significant if larval survival probability in nature is low. Under these circumstances, female fitness will be determined by the minimum number of offspring surviving to reproductive age, rather than by the maximum number hatching per clutch. Selection then favors females that "sacrifice" a portion of their clutches to sibling egg cannibalism, provided the fitness of surviving offspring is increased by doing so. Thus, sibling egg cannibalism represents a mechanism whereby females can trade offspring quantity for offspring quality.

The results of the first experiment suggest that adult coccinellid females can, and do, adjust the rate of sibling egg cannibalism in clutches by laying solitary eggs. Female *C. sanguinea* had the lowest fecundity of the three species on the *Ephestia* egg diet and also laid the largest proportion of their eggs singly (Fig. 1). Only a small proportion of the variation in number of eggs laid singly on a particular day was explained by clutch size, but the relationship was significant and convex in all species (Fig. 2), with the second order coefficients significant for both *H. axyridis* and *O. v-nigrum,* although not for *C. sanguinea.* Although obviously shaped by species-specific differences in fecundity, the parabolic inflection points were remarkably similar across species, indicating that the number of eggs laid singly did not increase with clutch size beyond a maximum of about six or seven eggs. Thus, females "protected" a proportion of their clutch from sibling cannibalism on days of low fecundity, but they tended to abandon this behavior on the days they produced large clutches. Infertile eggs were observed in all species and constitute another potential food source for cannibals (Hodek and Honek 1996), although in these experiments they desiccated before the eclosion of viable eggs, possibly as a result of low relative humidity in the laboratory.

Hatching synchrony was negatively correlated with cannibalism in all three species and explained as much as 24% of the variation (Fig. 4), making this another potential mechanism for maternal adjustment of cannibalism rate within clutches. The highest rate of sibling egg cannibalism occurred in *O. v-nigrum* clutches, together with the lowest survival rate for late-hatching larvae, and the lowest proportion of clutches without egg cannibalism (Fig. 3). We noted that *O. v-nigrum* larvae became active very quickly after hatching and searched rapidly for any unhatched eggs. The lowest proportion of eggs was cannibalized in *H. axyridis* clutches. Neonates of the latter species tended to remain on their empty eggshells for an extended period, affording late-eclosing eggs with more opportunity to hatch. Thus, differences in larval biology and behavior at eclosion account for some differences among species in rates of sibling egg cannibalism.

Because coccinellid larvae must typically travel some distance to find their first meal, the consumption of conspecific eggs by neonates before dispersal confers a substantial survival advantage (Banks 1956, Dixon 1959). Our hypothesis 4 was that egg cannibals would gain developmental advantages over noncannibals even when adequate food was available. In fact, egg cannibalism reduced the duration of the first instar in males and females of all three species, and in many cases this translated into a reduction in total developmental time. Both developmental time and body size are inferred to have considerable impact on coccinellid fitness (Dixon 2000). Faster development translates into earlier reproductive maturity and reduced exposure to predation, whereas increased adult weight means greater potential fecundity for females and various potential survival advantages associated with larger energy reserves.

Our hypothesis 5 was that females should benefit more from egg cannibalism than males, and it was supported for two of the three species in the second experiment. Females of both *H. axyridis* and *O. v-nigrum* had increased adult weights as a consequence of egg cannibalism, whereas differences in weight were not significant for males. This is consistent with an interpretation of females being more resource-sensitive than males, an idea that originates in sex allocation theory (Charnov 1982). Note that adult weight displayed a significant negative correlation with developmental time in both sexes of all three species, indicating that longer larval feeding times are not required to achieve larger body sizes in these beetles.

Balanced sex ratios were observed for both *C. sanguinea* and *H. axyridis* in the second experiment, but we obtained significantly more females than males for *O. v-nigrum.* Although the cause of this skew in sex ratio remains obscure, it seems unlikely to have arisen through the action of male-killing bacteria now known to commonly distort sex ratios in coccinellids and other insects (Jiggins et al. 2001). Indeed, no postzygotic mechanism seemed implicated, because neither the frequency of infertile eggs, nor larval mortality, was sufficiently high to account for the paucity of males.

In a study of egg cannibalism in *Coleomegilla maculata lengi* Timberlake, Gagne et al. (2002) showed that chemical cues were involved in egg-searching behavior by neonate larvae and demonstrated benefits in terms of developmental time and body weight in the second instar, although they did not follow insects to the adult stage and therefore could not discern sex-specific effects. Osawa (2002) performed a study with

H. axyridis that was very comparable to ours, but with one important difference: the larvae were reared on a diet of *Aphis spiraecola* Patch, an aphid species now known to be of inferior nutritional quality for *H. axyridis.* For example, Michaud (2000) found that *H. axyridis* adults weighed 45% more when reared as larvae on *Toxoptera citricida* (Kirkaldy) than when reared on *A. spiraecola* and that developmental time was almost halved. In the current study, *H. axyridis* noncannibal females weighed 68% more than did Osawa's noncannibal females, whereas males were twice as heavy, a strong indication that the *Ephestia* egg diet is far more suitable than *A. spiraecola* for larval development in this species, although genetic differences between source populations may also be involved. Similarly, our *H. axyridis* larvae pupated in just over 10 d, compared with almost a week longer for larvae in Osawa (2002), despite the fact that the latter were reared at a slightly higher temperature.

The one consistency between our study and that of Osawa (2002) is that egg cannibalism shortens the first instar, regardless of gender. In the study of Osawa (2002), cannibal males developed 4.2% faster than control males, compared with 2.5% faster in the current study, suggesting the benefits of egg cannibalism for males may be more pronounced when they feed subsequently on an inferior diet. However, Osawa's cannibal females failed to capture any benefits in terms of developmental time or adult weight on the *A. spiraecola* diet, whereas those reared on *Ephestia* eggs in the current study were 6.9% heavier and developed 4.2% faster than their noncannibal counterparts. Thus, *H. axyridis* females can actually benefit more from neonate egg cannibalism than males, in terms of both reduced developmental time and increased adult weight, but these benefits are contingent on access to a high quality diet for the balance of larval development.

Michaud (2003) studied rates of larval cannibalism and their developmental consequences in the same three species examined here and inferred that the high rates of larval cannibalism observed in *C. sanguinea* were at least partially due to the lower suitability of the *Ephestia* egg diet for this species relative to the other two. This inference was further supported by *C. sanguinea*'s faster larval development and superior survival on an exclusive diet of conspecific eggs relative to *Ephestia* eggs, effects not observed in either *H. axyridis* or *O. v-nigrum* (Michaud 2003). In the current study, noncannibal *C. sanguinea* larvae had significantly lower survival than their cannibalizing counterparts, but this was not true for the other two species. *C. sanguinea* was the only species in which males developed faster than females independent of cannibalism, and this probably reflects greater female sensitivity to the inferior diet represented by *Ephestia* eggs. Similarly, only *C. sanguinea* males benefited from egg cannibalism behavior in terms of accelerated development. In *O. v-nigrum*, for which *Ephestia* eggs represent a high-quality diet, female cannibals gained more than male cannibals in terms of both increased body size and faster development, although the reduction in developmental time for cannibal males might have also proven significant had not sample size been reduced by the female-biased sex ratio. The one explanation consistent with these results, collectively, is that the benefits of egg cannibalism behavior are potentially greater for female coccinellids than for males, but only if female larvae obtain a high-quality diet for the remainder of their development. The evolutionary and ecological implications of cannibalism behavior that yields gender-specific developmental benefits have yet to explored in this, or any other, group of insects.

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