

Reproductive Responses of Aphidophagous Lady Beetles (Coleoptera: Coccinellidae) to Nonaphid Diets: An Example from Alfalfa

DOUGLAS R. RICHARDS AND EDWARD W. EVANS

Department of Biology, Utah State University, Logan, UT 84322-5305

Ann. Entomol. Soc. Am. 91(5): 632-640 (1998)

ABSTRACT Aphidophagous lady beetles feed on a variety of prey in addition to preferred aphids. These alternative foods may serve only to maintain the predator but do not permit immature growth or adult reproduction. We tested whether larvae of the alfalfa weevil, *Hypera postica* (Gyllenhal), are alternative foods for adults of 2 species of lady beetles, *Coccinella septempunctata* L. and *C. transversoguttata* Brown, that breed in alfalfa while feeding on the larvae as well as aphids. When collected from alfalfa fields and placed on diets of weevils only, adults of both species generally gained weight. However, in contrast to females fed pea aphids, *Acyrtosiphon pisum* (Harris), females fed weevils produced very few eggs, almost all of which were laid in the first few days after the predators were placed on the weevil diet. When dissected, females fed weevils had only very small (previtellogenic) eggs in their ovaries. Similarly, females produced very few eggs, and held only previtellogenic eggs of very small size in their ovaries, when provided only sucrose (dissolved in water in 15% solution). When provided both weevils and sucrose, however, females of both species laid eggs in modest numbers. These results indicate that weevil larvae serve as alternative foods for the lady beetles, and that reproduction nevertheless can occur in the absence of preferred aphid prey when weevil larvae are combined in the diet with sucrose, another alternative food. Because nectar can serve as a natural source of sucrose in or near alfalfa fields, some reproduction by lady beetles may occur in fields infested with weevil larvae even in the absence or low abundance of aphids.

KEY WORDS *Coccinella septempunctata*, *Hypera postica*, alfalfa, biological control, oviposition, reproduction

LADY BEETLES OF the tribe Coccinellini (subfamily Coccinellinae) are distinctive in that most species of the tribe consume aphids as preferred prey (Gordon 1985, Hodek and Honek 1996). Many of these aphidophagous predators, however, are not restricted to eating aphids; they also attack various other insects and consume pollen and nectar (Hagen 1987, Pemberton and Vandenberg 1993, Hodek and Honek 1996). As elsewhere in North America (e.g., Elliott and Kieckhefer 1990, Neuenschwander et al. 1975, Frazer and Gill 1976, Wheeler 1977), aphidophagous lady beetles occur in high numbers in alfalfa fields of the Intermountain West (e.g., Evans and Youssef 1992). Adults returning from overwintering sites each spring feed heavily on pea aphids, *Acyrtosiphon pisum* (Harris) (Homoptera: Aphididae) (Davis et al. 1976), but also forage frequently at flowers of dandelions and other weeds growing nearby (personal observations). Further broadening of their diet results from these predators' frequent consumption of another abundant insect pest of alfalfa, the larval stage of the alfalfa weevil, *Hypera postica* (Gyllenhal) (Coleoptera: Curculionidae) (e.g., Webster 1911, Essig and Michelbacher 1933, Yakhontov 1934, Evans and England 1996).

The potential for lady beetle adults to contribute to biological control of alfalfa weevil larvae has received some attention over the years (e.g., Yakhontov 1938, Hussain 1975, Ouayogode and Davis 1981, Evans and

England 1996). Yet to be addressed by experiment, however, is the significance of consumption of weevil larvae for reproduction by lady beetles. Hodek and Honek (1996) have stressed the considerable variation in prey suitability for lady beetles, and distinguish between essential foods (those that support both larval development with high survival and subsequent reproduction by adults when serving as the sole source of nutrition) and alternative foods (those that do not support complete development or reproduction, but instead serve as sources of energy and nutrients to maintain the beetles in the absence of essential foods).

We present results of laboratory experiments conducted to address the suitability of alfalfa weevil larvae as prey in support of egg production and oviposition by lady beetles in alfalfa in the Intermountain West. Our experiments were conducted with 2 abundant lady beetle species that breed in this habitat while consuming weevil larvae as well as aphids—*C. septempunctata* L., an Old World species that recently has become established throughout North America (e.g., Obyrcki et al. 1987, Schaefer et al. 1987, Elliott et al. 1996); and *Coccinella transversoguttata* Brown, a similarly sized indigenous species. In addition to supplying the predators with alfalfa weevils and pea aphids, we also provided a source of sugar (sucrose dissolved in water) in particular treatments in some experiments, to mimic plant nectar upon which these pred-

ators also feed when breeding in alfalfa fields. Because the predators belong to the primarily aphidophagous Coccinellini, we followed Hodek (1970) in hypothesizing that alfalfa weevil larvae serve as alternative foods for the lady beetles (in the sense of being incapable of supporting reproduction when constituting the sole item in the diet) in contrast to essential foods as exemplified by pea aphids.

Materials and Methods

Methods Common to all Experiments. For all but the 6th experiment (see below), individuals of *C. septempunctata* and *C. transversoguttata* used as experimental subjects were collected as adults from alfalfa fields and adjacent vegetation near Logan, UT (see Richards [1997] for details). Prey for these adults (pea aphids and alfalfa weevil larvae) also were collected from these fields (aphids were collected daily and used immediately, whereas weevil larvae were collected at intervals and refrigerated until needed). Upon collection, adult lady beetles were refrigerated (and provided water only) for ≤ 19 d before being used in an experiment (the beetles were refrigerated to prevent them from aging too rapidly before an experiment could be conducted).

During experiments, pairs of conspecifics (female and male) were maintained individually in petri dishes (9 cm diameter) in an incubator at 25°C and a photoperiod of 14:10 (L:D) h. All pairs were provided a cotton-stoppered vial of water. According to experimental treatment, individual pairs also were provided aphids, weevil larvae, or sugar (sucrose) dissolved in the water. Consumption of prey by the lady beetles was quantified by determining the number of weevil larvae or aphids (or both) eaten over a 24-h period at intervals throughout experiments.

Egg production by females was quantified by checking petri dishes daily at 1100 and 1700 hours. Eggs were counted and then removed at each check. Because adult beetles may cannibalize their own eggs, petri dishes also were checked for eggs at other, less regular times throughout the day. On those few occasions when cannibalism did occur between checks, cannibalized eggs were counted and included from their chewed remains or the yellow spot they left at the site of attachment to the petri dish surface.

Female lady beetles that died during experiments were not replaced (dead males were replaced, however, so that females remained paired with a live male throughout an experiment). Overall, survivorship was high (70–100%) during experiments, and did not vary significantly within experiments among treatments and species (Richards 1997). Here we compare data for egg production and oviposition frequency only for those females that survived the entire experimental period.

Females were frozen and placed in 70% ethyl alcohol at the end of the experiment (or on the day they died during the experiment); they were then dissected to examine the ovaries. The 3 largest (most mature) follicles were used to determine reproductive condi-

tion; these were measured for length and width using a SZH Olympus stereomicroscope at 64 \times magnification. Follicular volume (size) was then approximated from the formula for a prolate spheroid, $(4/3)\pi ab^2$, where a is equal to half the follicle length and b is equal to half the follicle width (Phoofolo et al. 1995); mean volume of the 3 largest follicles was computed for each female. Females also were categorized as to their reproductive condition, the 3 most advanced follicles classed either as previtellogenic (white and <0.0035 mm³), vitellogenic (yellow but without a chorion, and 0.0035–0.0954 mm³), or postvitellogenic (yellow with a chorion, and >0.0954 mm³) (size criteria were developed independently of those of Phoofolo et al. [1995], but are very similar).

Reproduction on Diets of Weevils and Aphids. In the 1st experiment, prey were provided in excess, and paired adults of *C. septempunctata* and *C. transversoguttata* were randomly assigned to receive 1 of 2 experimental diets—pea aphids only (a daily ration of 60–80 4th instars), or alfalfa weevils only (a daily ration of 25 3rd instars). Many females were gravid, with swollen abdomens, at the time of collection (mid-July), and several eggs were laid in collecting vials before the beetles could be transferred on the same day to experimental petri dishes. Four to 7 pairs of adults of each species were maintained on each of the 2 diets for 10 d.

In the 2nd experiment, paired adults of *C. septempunctata* and *C. transversoguttata* (collected in mid-July, and refrigerated for 12 d) were assigned randomly to receive 1 of 3 diets: (1) a daily ration of 60–80 aphid 4th instars, (2) a daily ration of 25 weevil 3rd instars, or (3) a daily ration of 25 weevil 3rd instars and 6 aphid 4th instars. Five or 6 pairs of adults of each species were maintained on each of the 3 diets for 10 d.

Reproduction on Diets of Weevils and Sugar-Water. In the 3rd experiment, paired adults of *C. septempunctata* and *C. transversoguttata* (collected in early June and refrigerated for 4 d) were assigned randomly to receive 1 of 2 diets: (1) sugar-water only (provided as a 15% sucrose solution in the cotton-stoppered vial) or (2) sugar-water and a daily ration of 20 weevil 3rd instars. Fourteen pairs of *C. septempunctata* and 7 pairs of *C. transversoguttata* were maintained on each of the 2 diets for 10 d. At both the start and the conclusion of the experiment, all adults were weighed alive to the nearest 0.01 mg using a Sartorius Semimicro Balance.

In the 4th experiment, paired adults of *C. septempunctata* and *C. transversoguttata* (collected in mid-June and refrigerated ≤ 17 d) were assigned randomly to receive 1 of 3 diets: (1) sugar-water (15% sucrose solution), (2) a daily ration of 20 weevil 3rd instars, or (3) sugar-water and a daily supply of 20 weevil 3rd instars. Eighteen pairs of *C. septempunctata* and 12 pairs of *C. transversoguttata* were maintained on each of the 3 diets for 10 d. All experimental adults were weighed as described above at both the start and the conclusion of the experiment. An additional 12 females of each species were randomly selected at the

Table 1. Means and standard errors of the total number of eggs laid and the number of days on which eggs were laid by *C. septempunctata* and *transversoguttata* in Experiments 1, 2, 3, and 4

Experiment ^a	Diet	n		No. eggs		No. days	
		C7	Ct	C7	Ct	C7	Ct
1	Aphids	5	5	354.8 ± 31.2	385.4 ± 102.9	7.0 ± 0.5	7.8 ± 2.0
	Weevil	4	2	60.3 ± 25.2	37.5 ± 37.5	1.0 ± 0.4	1.5 ± 1.5
2	Aphids	5	6	193.6 ± 68.8	354.5 ± 37.0	6.6 ± 1.7	9.5 ± 0.3
	Weevil	4	4	0.5 ± 0.5	3.5 ± 1.4	0.5 ± 0.5	0.8 ± 0.3
	Weevil+6a	6	5	10.7 ± 4.3	40.0 ± 14.4	2.5 ± 0.6	3.6 ± 1.0
3	Sugar	14	6	5.5 ± 2.6	0	0.3 ± 0.1	0
	Sugar+w	12	8	107.8 ± 24.8	5.6 ± 4.8	3.8 ± 0.7	0.3 ± 0.3
4	Sugar	15	11	2.5 ± 1.4	5.9 ± 2.6	0.2 ± 0.1	0.4 ± 0.2
	Sugar+w	16	9	59.6 ± 14.8	28.7 ± 10.2	3.4 ± 0.6	2.0 ± 0.7
	Weevils	15	9	2.9 ± 2.2	0.7 ± 0.5	0.1 ± 0.1	0.2 ± 0.1

C7, *C. septempunctata*; Ct, *C. transversoguttata*. a, aphids; w, weevils.

^a Each experiment lasted for 10 d.

outset, frozen, and dissected to assess initial ovary condition.

In the 5th experiment, 30 pairs of *C. septempunctata* adults (collected in mid-July and refrigerated for 19 d) were assigned randomly to receive 1 of the same 3 diets used in the 4th experiment. At 2-d intervals over the next 10 d, 6 pairs of each treatment were chosen randomly to be dissected to assess ovary condition.

In the 6th experiment, adults of *C. septempunctata* were collected from sagebrush in mid-March upon recently emerging from hibernation sites (no aphids or other suitable prey were found). Twelve randomly selected females dissected upon collection had only undeveloped, previtellogenic follicles. The remaining beetles were refrigerated until early June, when an additional 15 females were dissected to determine reproductive condition. At this time, 15 pairs were randomly assigned to receive 1 of the same 3 diets used in the 4th and 5th experiments. The experiment was run for 25 d; all individuals were weighed both at the outset and at the conclusion of the experiment.

Voucher specimens of *C. septempunctata* and *C. transversoguttata* are deposited in the Utah State University Insect Collection.

Statistical Analyses. All experiments used completely randomized designs. Results were analyzed using SAS (SAS Institute 1993); 1- and 2-way analyses of variance (ANOVA) were performed using PROC ANOVA, or PROC GLM when the experimental design was unbalanced (unequal numbers of observations). Data for numbers of eggs laid, frequency of oviposition, and follicle size were log-transformed ($\ln[x+1]$) before analysis.

Results

Reproduction on Diets of Weevils and Aphids. *First Experiment.* When provided aphids only or weevils only (1st experiment), adults of *C. septempunctata* and *C. transversoguttata* readily ate prey in both diets, and did not differ in mean daily consumption rates for either diet; on average, individual pairs consumed 54 aphids each day (effect of predator species: $F = 0.04$; $df = 1, 8$; $P = 0.84$) or 12.5 weevil larvae ($F = 0.21$; $df = 1, 8$; $P = 0.66$). Females of both species provided a diet

of aphids laid many more eggs over the 10-d experimental period than did females provided a diet of weevils (Table 1; effect of diet: $F = 5.53$; $df = 1, 12$; $P = 0.036$; effect of predator species: $F = 0.77$; $df = 1, 12$; $P = 0.40$; interaction of diet and predator species: $F = 0.00$; $df = 1, 12$; $P = 0.95$). Females fed aphids also laid eggs on more days than did females fed weevils (Table 1; effect of diet: $F = 11.83$; $df = 1, 12$; $P = 0.005$; effect of predator species: $F = 0.02$; $df = 1, 12$; $P = 0.90$; interaction: $F = 0.10$; $df = 1, 12$; $P = 0.76$).

On a diet of aphids, females of both species laid eggs consistently throughout the experimental period, but on a diet of weevils they ceased laying eggs after the first few days (Fig. 1). When females were dissected, all individuals maintained on a diet of aphids had vitellogenic or postvitellogenic eggs, whereas with only 1 exception (a *C. transversoguttata* female), all individuals maintained on a diet of weevils had only previtellogenic eggs (Table 2). Correspondingly, the mean volume of primary follicles was significantly greater for females of both species maintained on a diet of aphids versus weevils (Table 2; effect of diet: $F = 15.79$; $df = 1, 20$; $P < 0.001$; effect of predator species: $F = 6.47$; $df = 1, 20$; $P = 0.019$; interaction: $F = 1.24$; $df = 1, 20$; $P = 0.28$).

Second Experiment. Similar results were obtained for females maintained on diets of aphids versus weevils in the 2nd experiment; females laid many eggs when provided aphids only, but almost no eggs when provided weevils only (Table 1). A 3rd group of females that was provided not only weevils but also 6 aphids per day (all of which were consumed by a female and her mate), produced relatively few eggs which were laid primarily in the 1st few days of the experiment (Fig. 2). In 2-way ANOVAS, there was no significant interaction between diet and predator species for either total number of eggs laid ($F = 0.11$; $df = 2, 24$; $P = 0.90$) or number of days on which a female laid eggs ($F = 0.32$; $df = 2, 24$; $P = 0.73$). The effect of diet was highly significant for both total number of eggs laid ($F = 26.76$; $df = 2, 24$; $P < 0.0001$) and number of days on which a female laid eggs ($F = 20.79$; $df = 2, 24$; $P < 0.0001$); *C. transversoguttata* females laid more eggs than did *C. septempunctata* females when maintained on a given diet ($F = 7, 19$; $df = 1, 24$; $P = 0.013$) but

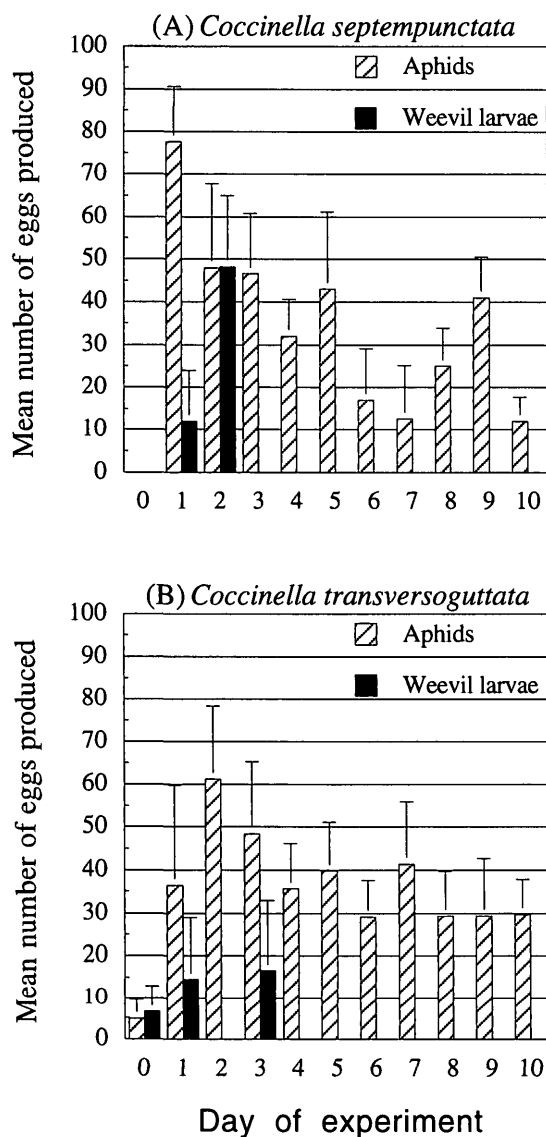


Fig. 1. Mean number of eggs (± 1 SE) produced each day (days 0–10; x-axis) by a female of (A) *C. septempunctata* and (B) *C. transversoguttata* when fed pea aphids or alfalfa weevil larvae (1st experiment).

did not lay on significantly more days ($F = 2.93$; $df = 1, 24$; $P = 0.10$).

Upon dissection, all individuals of *C. transversoguttata* and all but one of *C. septempunctata* maintained on a diet of only aphids had vitellogenic or postvitellogenic eggs, whereas none of the individuals maintained on a diet of only weevils did so (Table 2). Females maintained on a diet of both aphids and weevils were intermediate in reproductive condition; almost half of the females of both species had vitellogenic or postvitellogenic eggs. Correspondingly, follicle volume differed significantly with diet, and was particularly small for females of both species maintained on weevils only (Table 2; effect of diet: $F = 9.55$;

Table 2. Reproductive status of *C. septempunctata* and *C. transversoguttata* females maintained on varying diets, as reflected by the condition of the 3 most mature follicles in the ovaries at dissection, and by the mean volume of these follicles

Exp	Diet ^a	Follicular condition, no. females				Follicular volume ^a	
		C7		Ct		C7	Ct
		I	II	I	II		
1	Aphids	0	7	0	7	724 \pm 2.62	16.84 \pm 2.77
	Weevils	6	0	3	1	0.11 \pm 0.06	3.69 \pm 3.46
2	Aphids	1	4	0	6	8.26 \pm 2.90	14.97 \pm 2.33
	Weevils	6	0	5	0	0.20 \pm 0.06	0.30 \pm 0.11
	Both	3	3	4	2	6.60 \pm 4.65	3.48 \pm 2.16
3	S	14	0	7	0	0.22 \pm 0.04	0.05 \pm 0.02
	S \pm weevils	4	10	3	4	8.24 \pm 1.36	7.09 \pm 2.62
4	Initial ^b	1	10	2	4	11.54 \pm 1.46	11.68 \pm 4.38
	S	17	1	11	0	0.28 \pm 0.15	0.22 \pm 0.04
	Weevils	18	0	12	0	0.14 \pm 0.01	0.16 \pm 0.03
	S \pm weevils	5	13	4	8	7.17 \pm 1.84	8.95 \pm 2.90
5	S	30	0			0.06 \pm 0.01	
	Weevils	30	0			0.15 \pm 0.02	
	S \pm weevils	23	7			2.20 \pm 0.85	
6	Initial	15	0			0.08 \pm 0.01	
	S	15	0			0.04 \pm 0.01	
	Weevils	15	0			0.07 \pm 0.01	
	S \pm weevils	5	8			1.68 \pm 0.70	

See *Materials and Methods* for details of diet and follicular assessment. C7, *C. septempunctata*; Ct, *C. transversoguttata*. I, previtellogenic; II vitellogenic–postvitellogenic.

^a $\text{mm}^3 \times 100$. Both, both aphids and weevils; S, sugar-water.

^b Initial follicular condition at the outset of the experiment.

$df = 2, 28$; $P < 0.001$; effect of predator species: $F = 0.36$; $df = 1, 28$; $P = 0.55$; interaction: $F = 1.73$; $df = 2, 28$; $P = 0.20$).

Reproduction on Diets of Weevils and Sugar-Water. Third Experiment. When pairs of *C. septempunctata* and *C. transversoguttata* adults were provided weevil larvae daily plus sugar-water in the 3rd experiment, they consumed the larvae in large numbers (18.3 ± 0.5 and 15.4 ± 0.7 (mean \pm SE) larvae per day by *C. septempunctata* and *C. transversoguttata*, respectively; $F = 11.89$; $df = 1, 17$; $P = 0.003$). These adults increased substantially in weight over the course of the experiment, whereas adults maintained on a diet of only sugar-water changed relatively little in weight (Fig. 3). The effect of diet on weight change in females was highly significant ($F = 23.68$; $df = 1, 36$; $P < 0.0001$); females of *C. transversoguttata* gained significantly more weight during the experiment than did females of *C. septempunctata* ($F = 4.17$; $df = 1, 36$; $P = 0.049$; interaction of predator species and diet: $F = 0.17$; $df = 1, 36$; $P = 0.68$). The effect of diet also was highly significant for males ($F = 29.01$; $df = 1, 44$; $P < 0.0001$); males of the 2 species did not differ in weight gain ($F = 1.37$; $df = 1, 44$; $P = 0.25$; interaction of predator species and diet: $F = 0.02$; $df = 1, 44$; $P = 0.89$).

Females of *C. septempunctata* laid very few eggs (mostly on the 1st d) when maintained on a diet of sugar-water only, but laid eggs in moderate numbers throughout the experiment when also provided weevils (Table 1). Females of *C. transversoguttata* did not lay eggs when provided sugar-water only, and pro-

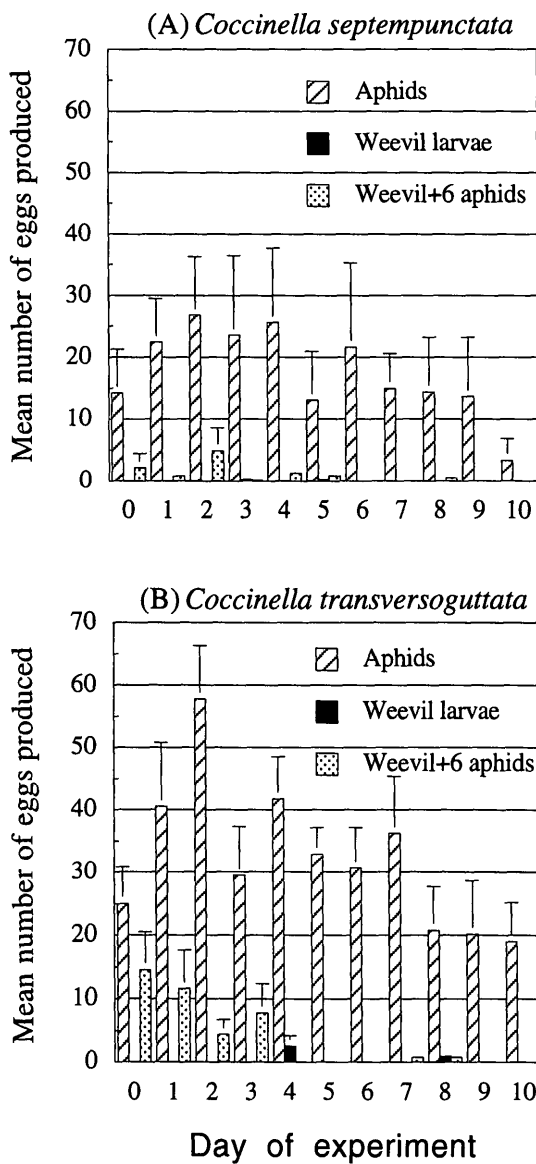


Fig. 2. Mean number of eggs (± 1 SE) produced each day (days 0-10; x-axis) by a female of (A) *C. septempunctata* and (B) *C. transversoguttata* when fed pea aphids, alfalfa weevil larvae, or alfalfa weevil larvae + 6 pea aphids daily (2nd experiment).

duced very few eggs (mostly on the 9th and 10th d of the experiment) when provided weevils as well as sugar-water (Table 1; total number of eggs laid, effect of diet: $F = 17.97$; $df = 1, 36$; $P < 0.0001$; effect of predator species: $F = 20.63$; $df = 1, 36$; $P < 0.0001$; interaction: $F = 7.31$; $df = 1, 36$; $P = 0.01$; number of days on which eggs were laid: effect of diet: $F = 20.31$; $df = 1, 36$; $P < 0.0001$; effect of predator species: $F = 24.13$; $df = 1, 36$; $P < 0.0001$; interaction: $F = 12.89$; $df = 1, 36$; $P = 0.001$).

Whereas many of the females of both species appeared gravid at the outset, none of either species had

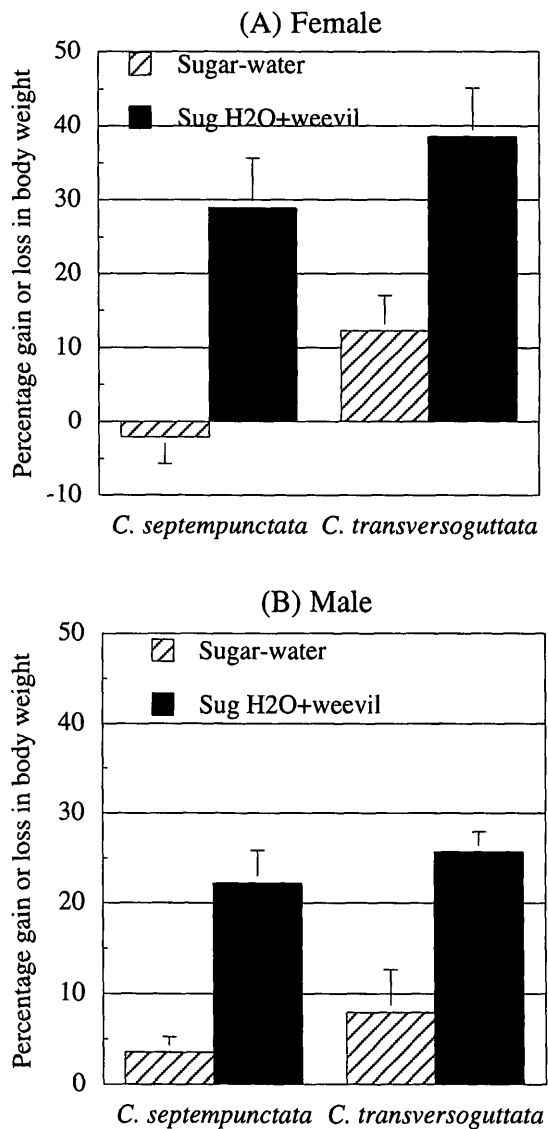


Fig. 3. Percentage gain or loss (mean ± 1 SE) in body weight of *C. septempunctata* and *C. transversoguttata* (A) females and (B) males over the 10-d experimental period when given a diet of sugar-water (15% sucrose) or sugar-water + 20 weevil 3rd instars (3rd experiment).

vitellogenic or postvitellogenic eggs after being maintained on a diet of sugar-water alone (Table 2). In contrast, 2/3 of females had such eggs when provided weevils as well as sugar-water. Correspondingly, follicle volume differed significantly with diet in females of both species (Table 2; effect of diet: $F = 33.09$; $df = 1, 38$; $P < 0.0001$; effect of predator species: $F = 0.28$; $df = 1, 38$; $P = 0.60$; interaction: $F = 0.15$; $df = 1, 38$; $P = 0.70$).

Fourth Experiment. The 4th experiment included a diet of only weevils along with the 2 diets (sugar-water only, and sugar-water plus weevils) compared in the 3rd experiment. A significant interaction between diet and predator species occurred in the number of weevil

larvae consumed daily by lady beetle pairs ($F = 6.21$; $df = 1, 45$; $P = 0.016$): *C. septempunctata* and *C. transversoguttata* pairs consumed similar numbers of weevils when provided only weevils (19.5 ± 0.3 and 19.2 ± 0.3 larvae per day), but *C. transversoguttata* pairs in particular consumed fewer weevils when also provided sugar-water (12.4 ± 1.9 versus 18.7 ± 0.4 larvae per day consumed by *C. septempunctata* pairs).

As in the 3rd experiment, adults provided only sugar-water differed from adults provided weevils in percentage change in weight over the course of the 4th experiment. When provided sugar-water only, females of both species lost considerable weight (Fig. 4). When provided weevils only or weevils plus sugar-water, *C. transversoguttata* females gained weight in equal and moderate amounts, whereas *C. septempunctata* females lost weight but lost less than on a diet of sugar-water only (effect of diet: $F = 9.27$; $df = 2, 69$; $P = 0.0003$; effect of predator species: $F = 6.27$; $df = 1, 69$; $P = 0.015$; interaction: $F = 2.35$; $df = 2, 69$; $P = 0.10$). Males of both species changed little in weight when provided sugar-water alone, and gained substantial amounts of weight when provided either weevils alone or weevils plus sugar-water (Fig. 4; effect of diet: $F = 17.12$; $df = 2, 74$; $P < 0.0001$; effect of predator species: $F = 0.31$; $df = 1, 74$; $P = 0.58$; interaction: $F = 0.89$; $df = 2, 74$; $P = 0.41$).

Females of both species laid eggs in moderate numbers throughout the experiment when provided weevils as well as sugar-water, but they laid very few eggs (on only the 1st or 2nd d) when provided only sugar-water or only weevils (Table 1; total number of eggs laid, effect of diet: $F = 22.52$; $df = 2, 69$; $P < 0.0001$; effect of predator species: $F = 0.30$; $df = 1, 69$; $P = 0.59$; interaction: $F = 1.68$; $df = 2, 69$; $P = 0.19$; number of days on which eggs were laid, effect of diet: $F = 27.06$; $df = 2, 69$; $P < 0.0001$; effect of predator species: $F = 0.39$; $df = 1, 69$; $P = 0.53$; interaction: $F = 1.92$; $df = 2, 69$; $P = 0.15$). Among females of both species dissected at the outset, or after being provided sugar-water and weevils for 10 d, most had vitellogenic or postvitellogenic eggs (Table 2). In contrast, all but one (*C. septempunctata*) female maintained on only sugar-water or only weevils had only previtellogenic eggs. Correspondingly, follicle volume differed significantly with diet for both species (effect of diet: $F = 25.00$; $df = 2, 98$; $P < 0.0001$; effect of predator species: $F = 0.10$; $df = 3, 98$; $P = 0.75$; interaction: $F = 0.17$; $df = 2, 98$; $P = 0.92$) and was especially large (close to initial condition) for females provided both sugar-water and weevils (Table 2).

Fifth Experiment. When maintained on the same 3 diets and dissected at 2-d intervals throughout the 5th experiment, *C. septempunctata* females consumed more weevil larvae when these were provided alone than when sugar-water also was provided (15.3 ± 0.6 versus 9.7 ± 0.9 larvae daily per pair; $F = 20.14$; $df = 1, 10$; $P = 0.0012$). Females laid no eggs during the experiment when provided sugar-water only or weevils only. Limited oviposition occurred when females were provided both sugar-water and weevils; 4 females laid a combined total of 106 eggs (13–22 eggs per

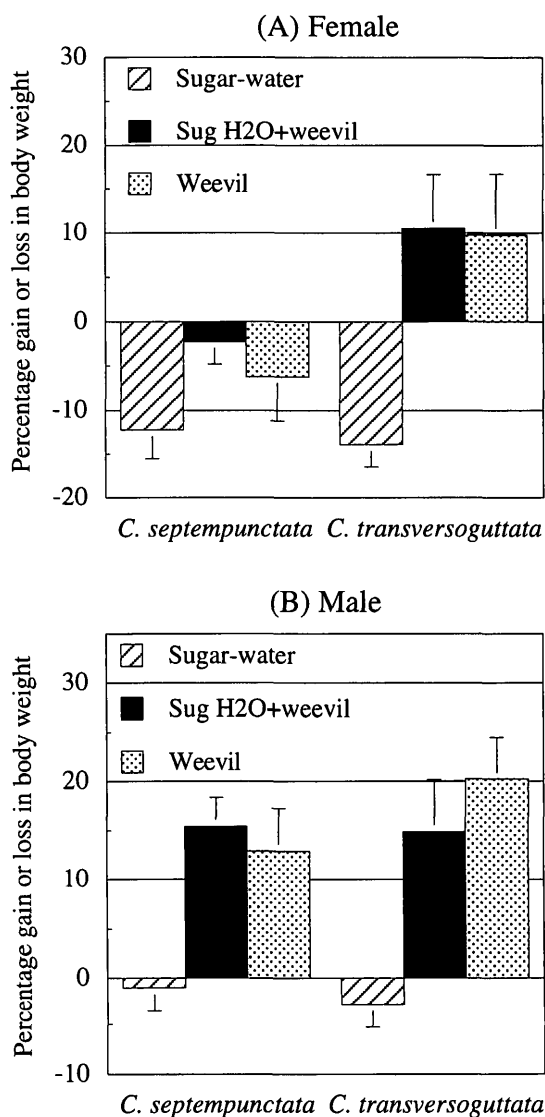


Fig. 4. Percentage gain or loss (mean \pm 1 SE) in body weight of *C. septempunctata* and *C. transversoguttata* (A) females and (B) males over the 10-d experimental period when given a diet of sugar-water (15% sucrose), sugar-water + 20 weevil 3rd instars, or 20 weevil 3rd instars (4th experiment).

day per female) throughout the 10-d experimental period. None of the females provided sugar-water only or weevils only, but nearly $\frac{1}{4}$ of the females provided both sugar-water and weevils, had vitellogenic or postvitellogenic eggs when dissected throughout the experimental period (Table 2). Correspondingly, follicle volume differed significantly with diet and remained relatively large or small according to diet throughout the experiment (Table 2; effect of diet: $F = 5.63$; $df = 2, 74$; $P = 0.0053$; effect of the day on which the female was killed: $F = 0.46$; $df = 4, 74$; $P = 0.76$; interaction of diet and day killed: $F = 0.53$; $df = 8, 74$; $P = 0.83$).

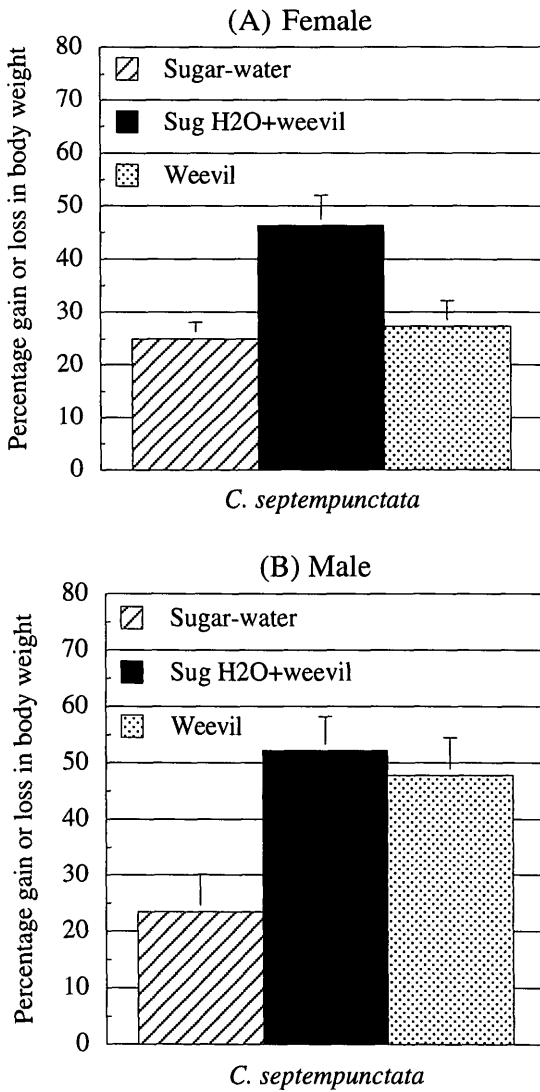


Fig. 5. Percentage gain or loss (mean \pm 1 SE) in body weight (over the 25-d experimental period) of (A) overwintered females and (B) males of *C. septempunctata* when given a diet of sugar-water (15% sucrose), sugar-water + 20 weevil 3rd instars, or 20 weevil 3rd instars (6th experiment).

Sixth Experiment. Overwintered adults of *C. septempunctata* again consumed more weevil larvae when these were provided alone than when sugar-water was also provided (19.4 ± 0.1 versus 17.8 ± 0.3 larvae daily per pair; $F = 29.39$; $df = 1, 18$; $P < 0.0001$). On average, both sexes gained weight over the 25-d experimental period on all 3 diets (Fig. 5). Weight gain was most pronounced for females provided sugar-water plus weevils ($F = 5.73$; $df = 2, 29$; $P = 0.008$ for effect of diet on weight gain), and for males provided weevils or sugar-water plus weevils ($F = 5.55$; $df = 2, 24$; $P = 0.01$). No eggs were laid by overwintered females that had been provided only sugar-water or only weevils, but females provided both sugar-water and weevils laid 38.6 ± 15.9 eggs (on 2.1 ± 0.9 d) during the

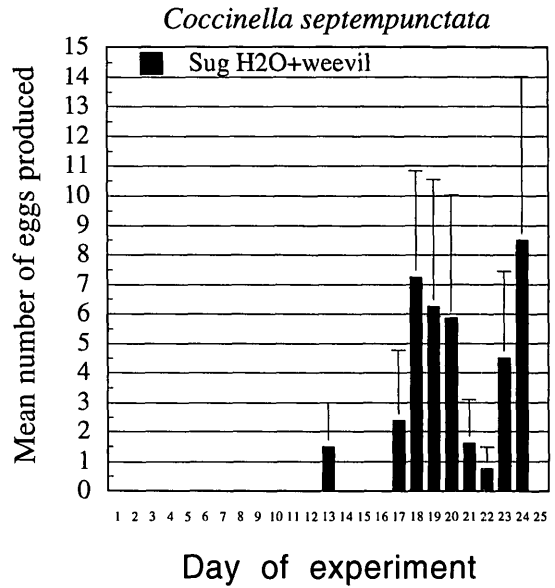


Fig. 6. Mean number of eggs (\pm 1 SE) produced each day (days 1-25; x-axis) by a female of *C. septempunctata* (C7) when given a diet of sugar-water (15% sucrose), sugar-water + 20 weevil 3rd instars, or 20 weevil 3rd instars (6th experiment).

experiment, with egg production starting on d 13 (Fig. 6). All the females dissected at the outset, or after 25 d on a diet of sugar-water only or weevils only, had only previtellogenic eggs, whereas in contrast, nearly $\frac{3}{5}$ of females provided both sugar-water and weevils had vitellogenic or postvitellogenic eggs (Table 2). Correspondingly, follicular volume differed significantly with diet, and was particularly large in females provided both sugar-water and weevils ($F = 6.38$; $df = 3, 54$; $P = 0.0009$).

Discussion

The wide range of foods in addition to aphids consumed in nature by aphidophagous lady beetles raises the obvious question of how important these foods are in the life cycles of these predators. Hodek (1973) and Hodek and Honek (1996) have emphasized the importance of assessing the suitability of these nonaphid accepted foods; they note that unlike an often surprisingly restricted set of aphids that serve as essential foods for a given lady beetle species, many alternative foods may prove incapable of supporting growth and reproduction when constituting the sole source of nutrition. Thus many aphidophagous lady beetles appear to be fairly specific in the foods that are suitable for their growth and reproduction; consumption of alternative foods by these predators appears only to sustain them temporarily in the absence of aphids that serve as essential foods.

The results of this study indicate that larvae of the alfalfa weevil serve as alternative foods for adults of *C. septempunctata* and *C. transversoguttata*. Survivorship

of adults fed weevils was high during experiments (overall 73% of females and 86% of males of both species combined survived for the full 10 d when fed only weevils in experiments (Richards 1997). Furthermore, in most cases the adults gained weight when maintained on a weevil diet. The 1 exception occurred in the 4th experiment, in which *C. septempunctata* females lost weight when fed weevils (with or without sugar-water). It is noteworthy that females lost even more weight in this experiment when provided only sugar-water. Perhaps the prolonged refrigeration before the experiment weakened the condition of all experimental females, resulting in weight loss for all treatments.

Although a diet of weevils was adequate to sustain and generally promote weight gain of *C. septempunctata* and *C. transversoguttata* adults, females appeared unable to produce eggs when fed solely on a diet of alfalfa weevil larvae. Almost all of the relatively few eggs that were laid by females maintained on a weevil diet appeared in the 1st few days of experiments, and hence were likely the products of previous consumption of aphids in the alfalfa fields from which these females were collected. In the 6th experiment, in which overwintered *C. septempunctata* females were used, no eggs were laid even after these females had fed on weevils for 25 d. In all experiments combined in which summer females were provided only weevils, 80 of 81 such females (99%) had only previtellogenic follicles in their ovaries when dissected; except for *C. transversoguttata* females in the 1st experiment, even the most mature follicles were very small (mean $\leq 0.003 \text{ mm}^3$). In contrast, experimental females of *C. septempunctata* and *C. transversoguttata* produced large numbers of eggs at frequent intervals when provided pea aphids exclusively, and 24 of 25 such females (96%) had postvitellogenic or vitellogenic follicles of large volume (mean $\geq 0.072 \text{ mm}^3$) when dissected (Richards 1997).

A diet of only sugar-water also sustained adult ladybirds, supporting high survivorship (but reduced weight gain in comparison with a diet of weevils). Overall, 92% of females and 96% of males of both species combined survived the full 10 d when maintained in experiments on sugar-water only (Richards 1997). However, just as when the predators were fed weevil larvae, so females of *C. septempunctata* and *C. transversoguttata* fed solely on sugar-water appeared unable to produce eggs. The few eggs laid by females maintained on the sugar-water diet appeared on the first few days of experiments, again suggesting that such eggs resulted from the lady beetles' consumption of aphids before collection. Of 95 females of both species maintained on only sugar-water in experiments, only 1 (*C. septempunctata*) female had vitellogenic follicles when dissected; all others had previtellogenic follicles of very small volume (mean $\leq 0.003 \text{ mm}^3$) (Richards 1997).

Hodek and Honek (1996) note that most researchers investigating the suitability of foods for aphidophagous lady beetles have not examined predator responses to combinations of foods (but see Phoofolo

and Obrycki 1997). In our experiments, intriguing results emerged when the 2 alternative foods, weevils and sugar-water, were provided simultaneously to *C. septempunctata* and *C. transversoguttata* adults. Provided with a mixed diet, females of *C. septempunctata* laid eggs throughout the 10-d experiments (note, however, that only small numbers of eggs were laid in the 5th experiment, perhaps both because the lady beetles were collected relatively late in the season and because they were refrigerated for almost 3 wk before being placed on experimental diets). Similar although less dramatic results occurred for *C. transversoguttata*. Upon dissection, a substantial number of females of both species had postvitellogenic or vitellogenic follicles of large size (mean $\geq 0.071 \text{ mm}^3$) in their ovaries (Richards 1997).

Ten-day experiments with mixed diets (weevil larvae plus sugar-water) were performed on summer females that were already reproductively active at the outset. Similar positive reproductive response to a mixed diet of sugar-water and weevil larvae also was observed in overwintered *C. septempunctata* females (6th experiment), which began laying eggs 13 d after being placed on experimental diets. At dissection, follicles of overwintered females on mixed diets were relatively large. In contrast, no eggs were laid over 25 d by females placed on diets of sugar-water alone or weevil larvae alone, and primary follicles remained small and undeveloped. Therefore, in *C. septempunctata*, the combination of 2 alternative foods in the predators' diet not only helped to maintain egg production but also stimulated its initiation after overwintering.

In summary, consumption of alfalfa weevil larvae by adults of *C. septempunctata* and *C. transversoguttata* appears to provide sufficient nutrition to sustain these predators in the absence or low abundance of aphid prey. Sole consumption of alfalfa weevil larvae, however, appears insufficient to support egg production and oviposition in these predators. It is quite intriguing that the predators will reproduce if a diet of weevil larvae is supplemented with sugar-water. This raises the possibility that in nature, even when aphids are absent or present in low numbers, these 2 species of lady beetles may be able to reproduce in alfalfa fields infested with weevil larvae if a suitable source of sugar (e.g., plant nectar or artificial honeydew) (Evans and Swallow 1993) is available. If so, this may be important in some cases for biological control of aphids by lady beetles. The predators may be able to increase in number through local reproduction even before aphid populations reach the relatively high levels that are necessary (when weevil larvae are not abundant) to induce lady beetle settlement and ovariole maturation (Honek 1978, 1980). Field studies are needed to assess whether indeed such is at times the case in alfalfa fields harboring weevils, aphids, and lady beetles.

Acknowledgments

We thank A. Stevenson, A. Mull, and L. Evans for collecting lady beetles in the field, F. Messina and J. Stark for

commenting on experimental design and results, and D. Sisson for advising on statistical procedures. Support was provided by the Utah Agricultural Experiment Station, NSF Grant No. DEB-9205721; and USDA Grant No. 95-37312-1664. Approved as Journal Paper No. 7002 of the Utah Agricultural Experiment Station.

References Cited

- Davis, D. W., G. E. Bohart, C. D. Griffin, B. A. Haws, G. F. Knowlton, and W. P. Nye. 1976. Insects and nematodes associated with alfalfa in Utah. *Utah Agric. Exp. Stn. Bull.* 494.
- Elliot, N. C., and R. W. Kieckhefer. 1990. A thirteen-year survey of the aphidophagous insects on alfalfa. *Prairie Nat.* 22: 87-96.
- Elliot, N. C., R. W. Kieckhefer, and W. Kauffman. 1996. Effects of an invading coccinellid on native coccinellids in an agricultural landscape. *Oecologia* 105: 537-544.
- Essig, E. O., and A. E. Michelbacher. 1933. The alfalfa weevil. *Calif. Agric. Exp. Stn. Bull.* 567.
- Evans, E. W., and S. England. 1996. Indirect interactions in biological control of insects: pests and natural enemies in alfalfa. *Ecol. Appl.* 6: 920-930.
- Evans, E. W., and J. C. Swallow. 1993. Numerical responses of natural enemies to artificial honeydew in Utah alfalfa. *Environ. Entomol.* 22: 1392-1401.
- Evans, E. W., and N. N. Youssef. 1992. Numerical responses of aphid predators to varying prey density among Utah alfalfa fields. *J. Kans. Entomol. Soc.* 65: 30-38.
- Frazer, B. D., and B. Gill. 1976. Coccinellids and aphids: a quantitative study of the impact of adult ladybirds (Coleoptera: Coccinellidae) preying on field populations of pea aphids (Homoptera: Aphididae). *J. Entomol. Soc. B.C.* 73: 33-56.
- Gordon, R. D. 1985. The Coccinellida (Coleoptera) of America north of Mexico. *J. N.Y. Entomol. Soc.* 93: 1-912.
- Hagen, K. S. 1987. Nutritional ecology of terrestrial insect predators, pp. 533-577. *In* F. Slansky and J. C. Rodriguez [eds.], *Nutritional ecology of insects, mites, spiders, and related invertebrates*. Wiley, New York.
- Hodek, I. 1970. Coccinellids and the modern pest management. *Bioscience* 20: 543-552.
1973. *Biology of Coccinellidae*. Publishing House of the Czechoslovak Academy of Science, Prague, Czechoslovakia.
- Hodek, I., and A. Honek. 1996. *Ecology of Coccinellidae*. Kluwer, Dordrecht, The Netherlands.
- Honek, A. 1978. Trophic regulation of postdiapause ovariole maturation in *Coccinella septempunctata* [Coleoptera: Coccinellidae]. *Entomophaga* 23: 213-216.
1980. Population density of aphids at the time of settling and ovariole maturation in *Coccinella septempunctata* (Col.: Coccinellidae). *Entomophaga* 25: 427-430.
- Hussain, M. 1975. Predators of the alfalfa weevil, *Hypera postica*, in western Nevada—a greenhouse study (Coleoptera: Curculionidae). *J. N.Y. Entomol. Soc.* 83: 226-228.
- Neuenschwander, P., K. S. Hagen, and R. F. Smith. 1975. Predation of aphids in California's alfalfa fields. *Hilgardia* 43: 53-78.
- Obrycki, J. J., W. C. Bailey, C. R. Stoltenow, B. Puttler, and C. E. Carlson. 1987. Recovery of the seven-spotted lady beetle *Coccinella septempunctata* (Coleoptera: Coccinellidae) in Iowa and Missouri. *J. Kans. Entomol. Soc.* 60: 584-588.
- Ouayogode, B. V., and D. W. Davis. 1981. Feeding by selected predators on alfalfa weevil larvae. *Environ. Entomol.* 10: 62-64.
- Pemberton, R. W., and N. J. Vandenberg. 1993. Extrafloral nectar feeding by ladybird beetles (Coleoptera: Coccinellidae). *Proc. Entomol. Soc. Wash.* 95: 139-151.
- Phoofolo, M. W., and J. J. Obrycki. 1997. Comparative prey suitability of *Ostrinia nubilalis* eggs and *Acyrtosiphon pisum* for *Coleomegilla maculata*. *Biol. Control* 9: 167-172.
- Phoofolo, M. W., J. J. Obrycki, and E. S. Krafusur. 1995. Temperature-dependent ovarian development in *Coccinella septempunctata* (Coleoptera: Coccinellidae). *Ann. Entomol. Soc. Am.* 88: 72-79.
- Richards, D. R. 1997. The role of alfalfa weevil larvae as prey in adult maintenance and reproduction by two ladybird beetle species. M.S. thesis, Utah State University, Logan.
- SAS Institute. 1993. *SAS/STAT user's guide: statistics, version 6*. SAS Institute, Cary, NC.
- Schaefer, P. W., R. J. Dysart, and H. B. Specht. 1987. North American distribution of *Coccinella septempunctata* (Coleoptera: Coccinellidae) and its mass appearance in coastal Delaware. *Environ. Entomol.* 16: 368-373.
- Webster, F. M. 1911. The alfalfa weevil. *U.S. Dep. Agric. Circ.* 137.
- Wheeler, A. G., Jr. 1977. Studies on the arthropod fauna of alfalfa. VII. Predaceous insects. *Can. Entomol.* 109: 423-427.
- Yakhontov, V. V. 1934. The alfalfa weevil *Phytonomus (Phytonomus) variabilis* Hbst.). *Rev. Appl. Entomol. Ser.* 22: 334-336.
1938. Practical results of an experiment. A biological method of controlling pests of lucerne and cotton. *Rev. Appl. Entomol. Ser. A* 26: 238-239.

Received for publication 13 January 1998; accepted 14 April 1998.