SOME EFFECTS OF POPULATION DENSITY ON THE LIFE HISTORY OF THE OBLIQUE-BANDED LEAFROLLER

by

Lisa M. Poirier

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Name: Lisa Marie Poirier

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Examining Committee:

Chairman: Dr. Peter Belton

Dr. J.H. Borden, Professor, Senior Supervisor

Dr. M. Mackauer, Professor

Dr. G. Judd, Agriculture Canada Research Station, Summerland, B.C., Public Examiner

Date Approved 17 August, 1989

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Some Effects of Population Density on the Life History of the Oblique-Banded Leafroller

Author:

(signature)

Lisa Marie Poirier

(name)

August 16/89

(date)

ABSTRACT

Larvae of the obliquebanded leafroller, *Choristoneura rosaceana* Harris, were reared on bean plants at two densities of 10 and 50 larvae per 15 plants in 24 x 24 x 24 cm cages. The host plants were replaced frequently so that food was apparently not a limiting factor. Larvae at the higher density exhibited an altered larval dispersal pattern, lower survival, smaller pupal size, and a longer pupal period. Since all of these effects could have contributed to reduced reproductive fitness for crowded insects, it was hypothesized that some form of spacing mechanism is active in this species. One such mechanism could be the dispersion of egg masses laid by adult females. Investigations into oviposition disclosed that laboratory-reared females deposit their egg masses in a significantly non-random, uniform manner. Experiments were conducted in oviposition chambers in which gravid females were exposed either to egg masses recently deposited by other females, or to waxed paper from which egg masses had been removed. These experiments demonstrated that females can detect and avoid egg masses laid by other females, and that this phenomenon is most likely due to an oviposition deterrent (epideictic) pheromone laid down with each egg mass.

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INTRODUCTION

The effects of population density on the life histories of insects have been widely documented (Peters and Barbosa 1977, Long 1953). More recently, researchers have become interested in the mechanisms by which insects detect, respond to, and avoid non-optimal densities (Prokopy *et al.* 1984, Prokopy 1981a). It is possible that such mechanisms could be exploited in integrated pest management (IPM) programs, either to reduce the actual pest population in an area, or to reduce the reproductive success of later generations. The information is also important for mass-rearing programs, in which densities frequently exceed those found in natural situations.

Orchard insect pests have been prime targets for the development of IPM programs. One of these is the obliquebanded leafroller, *Choristoneura rosaceana* Harris (Lepidoptera: Tortricidae). *C. rosaceana* has a broad host range, but prefers members of the family Rosaceae. It is a pest of numerous deciduous tree crops across North America, including apples (Reissig 1978), pistachios (Rice *et al.* 1988), and filberts (AliNiazee 1986). Although it is not usually a major pest, it can be difficult to control because of its prevalence on native vegetation. This fact and its close relationship to the eastern and western spruce budworms, *C. fumiferana* (Clem.) and *C. occidentalis* Free., both of which can devastate vast areas of forest land, make the obliquebanded leafroller an ideal candidate for preliminary studies.

This paper examines the effects of high population density on *C. rosaceana* larvae, and the possible existence of an epideictic, or density-regulating, pheromone in newly-laid egg masses.

EFFECT OF REARING DENSITY

1.0 INTRODUCTION

The impact of population density on insect biology has been widely researched for many years. Numerous parameters, such as survival, developmental rate, size, feeding behaviour, dispersive and aggregative behaviour, aggression, and physiology have been shown to be density dependent in some way (Peters and Barbosa 1977, Watt 1960, Long 1953). Responses to population density may increase in intensity as numbers increase, or vary outside an optimal density range. Thresholds have been indicated in several cases, below which there is no effect (Peters and Barbosa 1977). Frequently, responses are most pronounced in immature stages.

In order for density responses to be manifested, individual insects must be able to detect the density of conspecifics in a particular area. Several mechanisms for achieving this have been demonstrated in laboratory settings, the simplest of which entails the frequency of encounters with conspecifics. These encounters may involve physical contact (Moller 1988, Tschinkel and Willson 1971) or actual aggression (Mitchell 1980, Ankersmit and van der Meer 1973). Others have proposed that visual (Shapiro 1981, Rausher 1979) or acoustic cues (Doolan 1981) may also be important. The most frequently suggested mechanisms of density detection involve chemical communication. It has been proposed (Kidd 1977) that nymphs of the lime aphid, *Eucallipterus tiliae* (L.) receive information on population

density through the concentrations of certain salivary substances in a leaf. Sanders (1987) has shown that dispersal behaviour of mated female spruce budworm moths, *C. fumiferana*, increases when high concentrations of sex pheromone are present, implying a response to density. Larvae of the Mediterranean flour moth, *Ephestia kuehniella* Zeller, take longer to pupate at high or low population densities. This density response can be induced by exposure to very high or low concentrations, respectively, of a mandibular gland secretion from conspecific larvae (Corbet 1971). This secretion also deters oviposition in adult female flour moths (Corbet 1973). All of these examples suggest that insects can perceive chemical cues which provide information about population density, and that they are able to alter their behaviour accordingly.

Density effects such as those described above can have numerous practical implications. For example, reducing an insect's population density through some control measure might increase the survival, size, and fecundity of those remaining, thus actually aggravating the problem in the long term (Watt 1960). Negative responses to high population densities can pose problems in the mass-rearing of insects, whether for research or for the commercial production of biological control agents (Mackauer 1976). Perhaps most importantly, if negative responses could be induced artificially when population densities are not excessive, some measure of pest control might be achieved.

This study examines the impact of population density on larvae of the obliquebanded leafroller, *Choristoneura rosaceana*. The following life history parameters were considered: larval dispersal, survival, times to pupation and to eclosion, and pupal size. The experiments were designed to mimic a standard rearing procedure closely, in order to make the results as applicable as possible to the laboratory colony from which the test insects were taken.

2.0 MATERIALS AND METHODS

2.1 REARING OF STOCK COLONY

All insects were taken from a four-year-old colony of *C. rosaceana* maintained at 24 to 26°C, and approximately 50 to 60% relative humidity (R.H.). The lighting regime was 18:6 h L:D.

Larvae were reared on broad beans planted in greenhouse trays. They were transferred to fresh plants whenever the foliage became depleted. Since larvae frequently wandered from the trays just prior to transfers, fresh plants were kept nearby to intercept them. Wanderers which did not find the fresh beans on their own were placed on new plants by hand.

Pupae were collected from the plants every 1 to 2 days and placed in paper bags for eclosion, mating, and oviposition. Twenty to 30 unsexed pupae were kept in each bag. When more than 10 egg masses had accumulated in a bag, the remaining pupae and moths were placed in a new bag or were used to increase the populations in existing bags.

Small triangles of paper with egg masses attached were cut from the bags and placed on moist filter paper in petri dishes. When the eggs reached the "black-headed stage", with larval head capsules visible through the chorions, these triangles were pinned to fresh bean leaves. Larvae emerged within 1 to 2 days, directly onto the beans.

2.2 EXPERIMENTAL PROCEDURE

The methodology used in this experiment was adapted from the standard rearing procedure. The temperature, R.H., and lighting conditions were similar for the rearing and experimental rooms. Similar rearing techniques and materials were utilized.

Trays of healthy, 2-week-old bean plants were divided into halves, each containing 15 plants. Both halves were infested on the same day with newly hatched larvae of *C. rosaceana*. One half received 10 larvae, while the other half received 50. The larvae were taken from different egg masses, to minimize any effects of genetic homogeneity which might have influenced the insects' response to crowding (Bryant and Sokal 1967).

Both halves of the trays were covered with 24 x 24 x 24 cm plexiglass cages pushed into the soil to a depth of approximately 0.5 cm to prevent larval escapes and to minimize air movement between cages. The trays were rotated 180° every second day to ensure an equivalent light intensity regime for the two groups. The temperature and R.H. inside the cages were not quantified, but both high and low density treatments would have been affected in the same manner.

The trays were checked daily between 1200 and 1300 h, and transfers to new beans were made while green leaves were still present in both density treatments. Larvae in both halves of a tray were always transferred on the same day. The low density

half was opened for transfers or data collection prior to the high density half to reduce the possibility of odour contamination between treatments. When present, pupae were removed from the cages daily, sexed, and placed in paper bags labelled with appropriate tray and group numbers. These bags were checked daily for eclosion.

Both numbers and instars of larvae found "wandering" on the plexiglass were recorded. The instar was estimated from larval size, based on information from the stock colony. Insects which had attached their leafrolls to the cage, and those which were attempting to pupate on the plexiglass were not included. As in the normal rearing procedure, wanderers were returned to the plants. The number of larvae remaining in each cage was noted on transfer dates. Ideally, this information would have been obtained more frequently and regularly. However, the larvae were considerably agitated by the counting procedure, and the plants sustained serious physical damage. This was particularly true when small, tunnelling larvae were present.

The sizes of new pupae were determined daily. Length was used as an index of weight, because it could be measured more quickly and involved less handling of the pupae. Each pupa was sexed according to Robertson's (1985) criteria indicating five visible abdominal sternites in males and four in females. Each day, all newly-emerged moths were sexed on the basis of the presence (males) or absence (females) of tufts of hair at the apex of the abdomen (Robertson 1985).

Ten replicates of the two treatments were completed over a 12-month period.

2.3 STATISTICAL ANALYSES

t-Tests (Steel and Torrie 1980) were used to compare the mean percentages of insects surviving to adulthood from hatching and from pupation, and the mean pupal lengths for males and females. The Kolmogorov-Smirnov two-sample test was used to compare cumulative frequency distributions of numbers pupating and eclosing to adulthood over time (Steel and Torrie 1980). Within treatments, a Student-Newman-Keuls' multiple comparison analysis (Steel and Torrie 1980) was used to determine differences in the numbers of wandering larvae between instars. The mean numbers of larvae wandering per cage at set times from hatching were compared graphically. These data were transformed logarithmically (Steel and Torrie 1980) prior to calculation of standard errors.

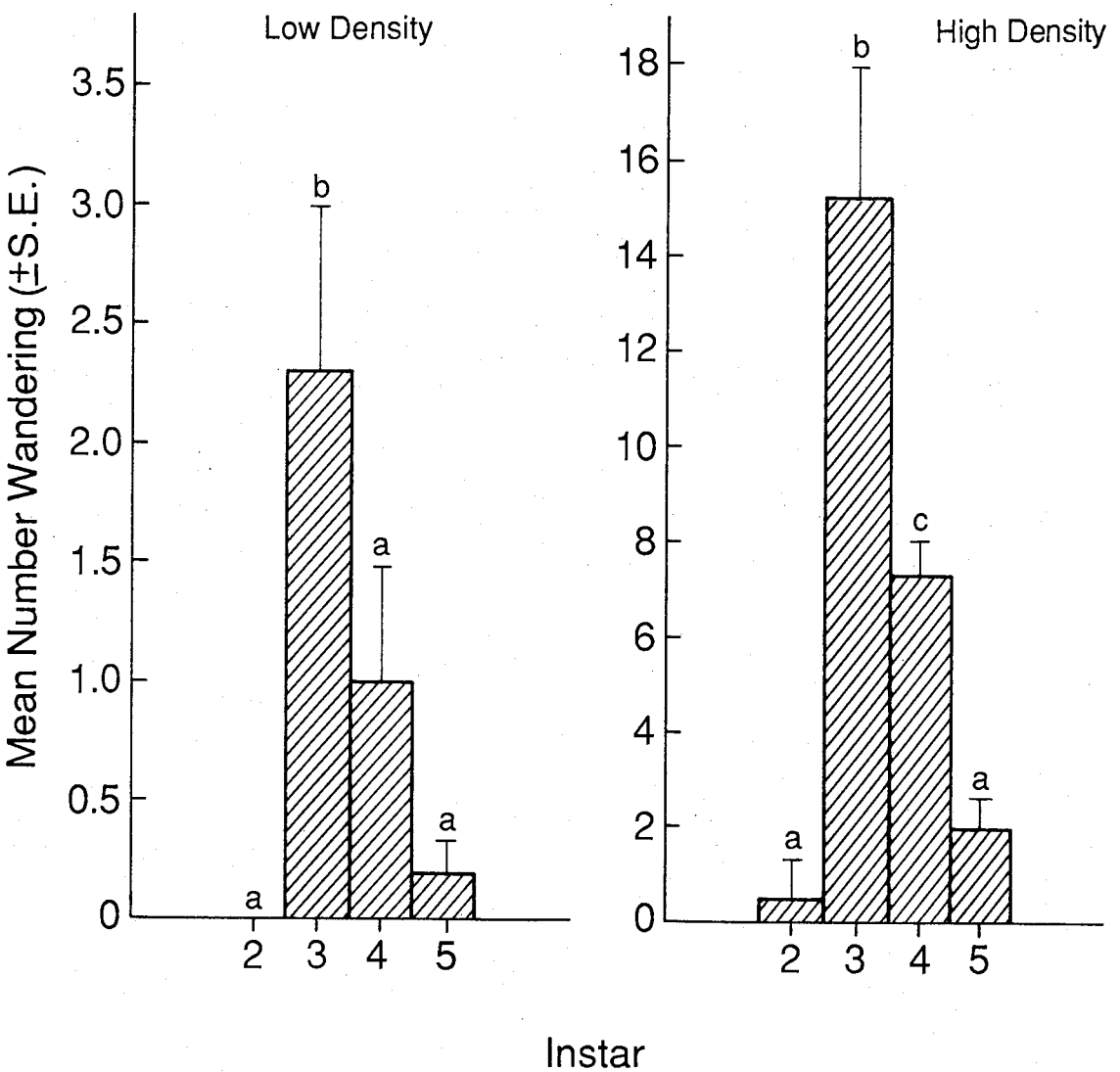
3.0 RESULTS AND DISCUSSION

3.1 WANDERING LARVAE

In both high and low density treatments, a significant majority of the "wandering" larvae were in the third instar (Fig. 1). Second, fourth, and fifth instar larvae wandered less frequently. However, in the high density treatment, fourth instar larvae also wandered more frequently than did second or fifth. No first instar larvae, and very few second instar larvae were observed attempting to disperse. In a natural situation, these very young larvae could be subject to quick starvation and dessication if they did not encounter a new host plant within a very short time. This problem exists for other lepidopterans (Jennings *et al.* 1983, Dethier 1959). Due to the nature of this experimental system, other explanations are also possible. The lack of air currents inside the cages may have reduced the incidence of dispersal on silk threads (Nealis and Regniere 1987) which is common in early-instar tortricid larvae. However, the abundant bean foliage could have provided enough food and space for the small larvae at both densities, so that long range dispersal was not necessary.

Although the two treatments were not compared directly, an upper size limit for dispersal is also implied, which may be based on the degree of crowding. By the time the later larval stages are reached, the larval density could have been reduced

Fig. 1. Mean numbers of wandering *C. rosaceana* larvae per cage for each instar at high (50 larvae per cage) and low (10 larvae per cage) densities on beans replaced frequently. n=10. Bars with same letter above are not significantly different, Student-Neuman-Keuls test, $P<0.05$.

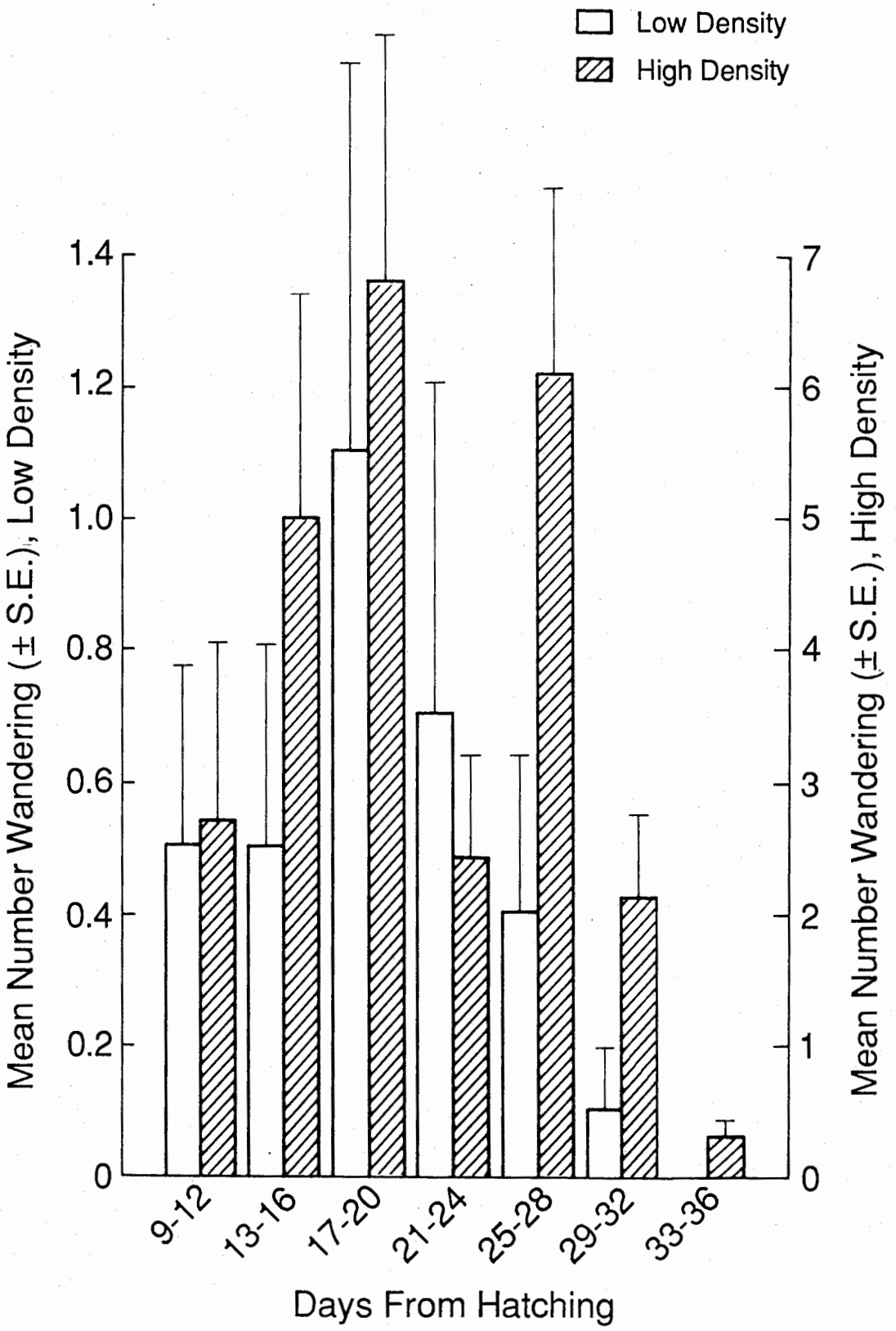

Low Density
High Density
Mean Number Wandering (±S.E.)
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
18
16
14
12
10
8
6
4
2
0
a
b
a
a
a
b
c
a
2
3
4
5
Instar

by those that have pupated, died, or (in a natural population) dispersed out of the area. Remaining larvae would not need to leave if the food supply is maintained. More persistent crowding in the high density treatment would explain why the high dispersal rate continued into the fourth instar (Fig. 1).

Fig. 2 shows the mean number of larvae wandering per cage over time for both density treatments. More wanderers appear to have been present in the high density treatment between 25 and 32 days after hatching, near the end of the last larval stage. It was not possible to test these differences statistically, since individual observations were not independent. Ankersmit and van der Meer (1973) found that *Adoxophyes orana* F.R. larvae frequently exhibited retarded growth under crowded conditions. My results could be due to large numbers of such suppressed larvae in the high density treatment reaching a suitable size for dispersal later in the developmental period. Enough competitors would have died or pupated by this point that these suppressed larvae could begin to grow. However, there could still be enough competitors left to induce wandering (Peters and Barbosa 1977), if such wandering is induced by conspecifics. Nealis and Regniere (1987), Corbet (1971), and Hodjat (1970) have all demonstrated increased larval dispersal tendencies under crowded rearing conditions.

The results in Figs. 1 and 2 suggest that density may have an important influence on larval dispersal by *C. rosaceana*. The trigger is unknown, but was probably hunger. There were still

Fig. 2. Mean numbers of wandering *C. rosaceana* larvae per cage over time at high (50 larvae per cage) and low (10 larvae per cage) densities on beans replaced frequently. n=10. Numbers for high density treatment coded by dividing by 5 prior to calculations.


Low Density
High Density
Mean Number Wandering (± S.E.), Low Density
Mean Number Wandering (± S.E.), High Density
0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
0
1
2
3
4
5
6
7
9-12
13-16
17-20
21-24
25-28
29-32
33-36
Days From Hatching

green, unoccupied leaves present at each transfer date, but these often had silk on the leaf surface or were in close proximity to occupied leaf rolls. They may, therefore, have been perceived as uninhabitable by the larvae. It is also possible that these leaves were unpalatable for some other reason, such as induced defenses in the host plants (Rhoades 1985).

3.2 SURVIVAL

The percentage of insects surviving to pupation was 72.0% in the low density treatment and 23.4% in the high density treatment (Fig. 3). If such significant mortality occurs in nature at high densities, it would evidently be of adaptive advantage for *C. rosaceana* to develop mechanisms to detect and avoid larval crowding. Use of oviposition pheromones (Prokopy 1981a), larval aggression (Ankersmit and van der Meer 1973), and larval epideictic pheromones (Corbet 1971) are some potential mechanisms which might be adopted. If greater mortality occurs at high densities after the third instar (Fig. 3), when larvae are capable of dispersing from the crowded area, selection pressure towards development of dispersal-inducing systems will be even greater (Prokopy 1981a).

Almost four times more larvae survived to adulthood in the low than in the high density treatment (Table 1). However, this difference was not evident in the percentage of pupae surviving to adulthood, which did not appear to be density-dependent. The

Fig. 3. Mean percent of *C. rosaceana* larvae surviving to pupation over time at high (50 larvae per cage) and low (10 larvae per cage) densities. Larvae reared on bean plants replaced frequently. n=10.

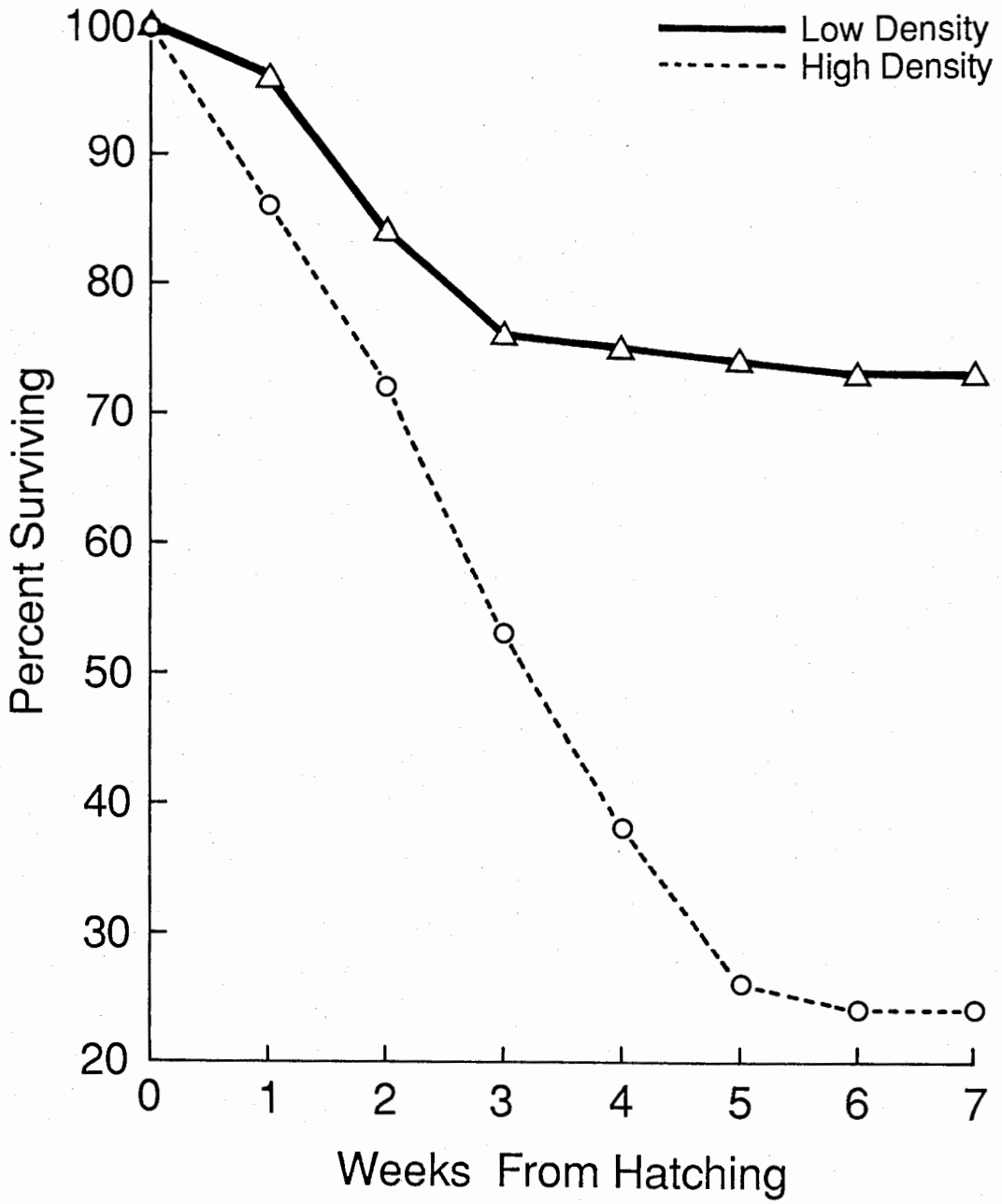

Low Density
High Density
Percent Surviving
100
90
80
70
60
50
40
30
20
0
1
2
3
4
5
6
7
Weeks From Hatching

Table 1. Comparison of mean percent of *C. rosaceana* surviving to adulthood in high (50 larvae per cage) and low (10 larvae per cage) density treatments over all instars, and over pupal instar only.

	All instars		Pupal instar	
Treatment	No. larvae	% surviving to adulthood ($\bar{x}$ ± S.E.)[a]	No. pupae	% surviving to adulthood ($\bar{x}$ ± S.E.)[a]
Low density	100	48.0 ± 2.5 a	72	68.6 ± 4.6 a
High density	500	13.6 ± 1.1 b	117	61.3 ± 5.2 a

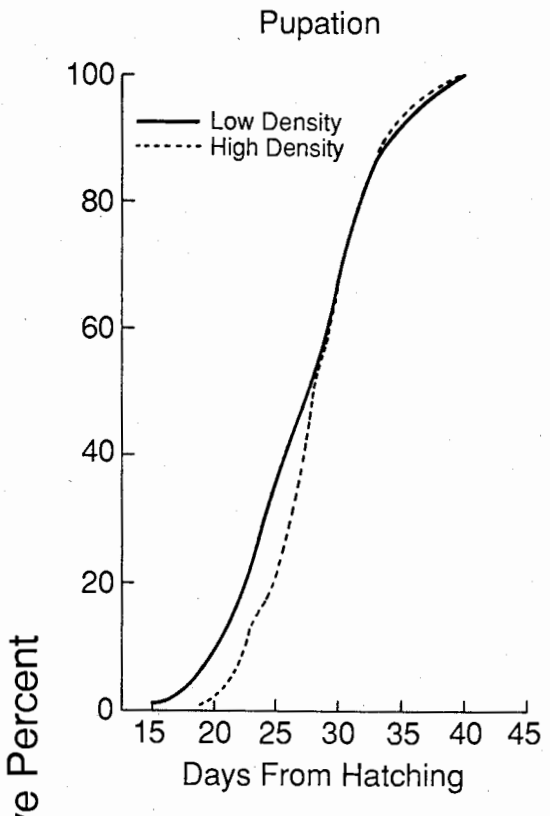
[a] Means in a column followed by same letter are not significantly different, t-test, $P<0.05$.

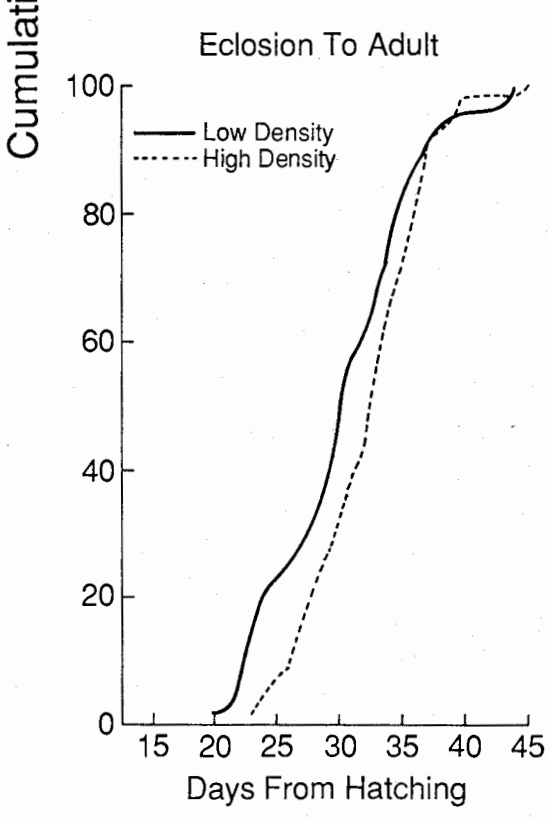
design of this experiment did not allow observations of which pupae survived and which did not. These results can be accounted for by the differential survival to pupation (Fig. 3), which has been found in several other insects, including *Ephestia kuehniella* Z. (Cotter 1974), and *Lymantria dispar* (L.) (Leonard 1968). Again, in a natural system, selection would favour the development of density detection and avoidance mechanisms which are operative either very early in the life cycle before larvae become established (Prokopy 1981a), or after they reach the dispersive third instar as differential mortality becomes pronounced (Fig. 3).

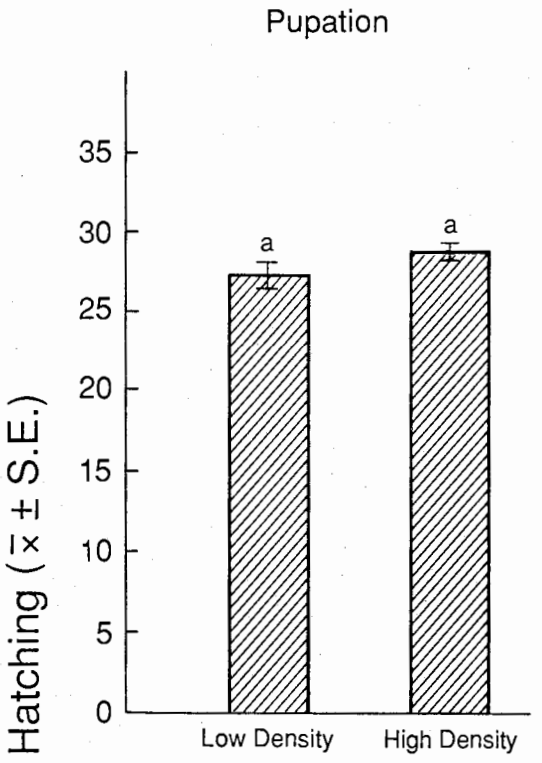
3.3 TIME TO PUPATION AND ECLOSION

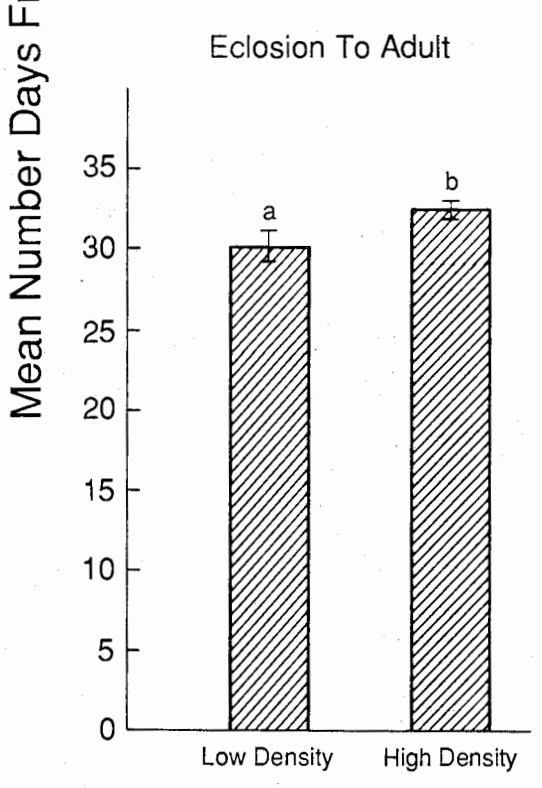
No significant differences were found between the two densities in the cumulative percentages of surviving insects pupating over time (Fig. 4) (Kolmogorov-Smirnov two-sample test, $P>0.05$). As well, there was no difference between the mean times to pupation for the two treatments. However, there was significantly faster development and a shorter duration from hatching to adulthood in the low than in the high density treatment, based on the same statistical tests (Fig. 4). These results indicate that the insects from the high density treatment spent longer in the pupal stage than those from the low density treatment. Similar results have been reported for *Culex nigripalpus* Theobald (Peters and Barbosa 1977) and *Spodoptera littoralis* (Boisd.) (Hodjat 1970).

Fig. 4. Comparison of the rate of, and mean duration to pupation and eclosion for *C. rosaceana* reared at high (50 larvae per cage) and low (10 larvae per cage) densities on bean plants replaced frequently. Different letters above paired bars indicates significant difference, t-test, $P<0.05$.


Pupation
Cumulative Percent
Low Density
High Density
100
80
60
40
20
0
15 20 25 30 35 40 45
Days From Hatching


Eclosion To Adult
Low Density
High Density
100
80
60
40
20
0
15 20 25 30 35 40 45
Days From Hatching


Pupation
Mean Number Days From Hatching (x̄ ± S.E.)
35
30
25
20
15
10
5
0
a
a
Low Density
High Density


Eclosion To Adult
35
30
25
20
15
10
5
0
a
b
Low Density
High Density

3.4 PUPAL SIZE

Pupae of each sex from the low density treatment were significantly longer than pupae of the same sex from the high density treatment (Table 2). This is a common result in studies of this nature (Corbet 1971, Hodjat 1970, Leonard 1968). In both treatments female pupae were found to be significantly longer than male pupae. Since insects from the high density treatment are smaller, it is also possible that they have lower wing loading, which, in nature, could improve their ability to disperse and search for new hosts (Sanders and Lucuik 1975). They would probably also have reduced fecundity (Corbet 1971, Leonard 1968), and, therefore, be less fit as measured by reproductive success.

Table 2. Comparison of mean pupal lengths (mm) for male and female *C. rosaceana* reared at high (50 larvae per cage) and low (10 larvae per cage) densities.

	Males		Females	
Treatment	No. pupae	Length (x ± S.E.)[a]	No. pupae	Length (x ± S.E.)[a]
Low density	24	10.7 ± 0.2 a	29	11.3 ± 0.2 a
High density	42	10.2 ± 0.1 b	31	10.9 ± 0.1 b

[a] Means in a column followed by same letter are not significantly different, t-test, $P<0.05$. At each density, female pupae were larger than males, t-test, $P<0.05$.

OVIPOSITION BEHAVIOUR

1.0 INTRODUCTION

As discussed previously, density can have far-reaching impacts on insect population biology. When these impacts are detrimental to individual fitness, i.e. reproductive success, mechanisms are likely to have evolved for the detection and avoidance of non-optimal densities. Those individuals which have developed and maintained such mechanisms will contribute proportionately more to the gene pool, and the mechanisms will become widespread in the population (Prokopy *et al.* 1984, Prokopy 1981a).

I have already shown that high population density can have detrimental effects on larval *C. rosaceana* which persist throughout their lifespan and may reduce individual fitness. Assuming that such effects also occur in nature, it is logical to hypothesize that some mechanism exists for preventing overcrowding of the larvae. Although larval dispersal may be important, it is extremely risky and, in numerous species, it is a major cause of mortality (Jennings *et al.* 1983, Dethier 1959). One possible way to minimize the problems of both overcrowding and larval dispersal is for the female moth to avoid conspecific egg masses when ovipositing. This phenomenon requires that she have a method for detecting the presence of such egg masses, and that she can adapt her oviposition behaviour accordingly.

Numerous insects, several of which are lepidopterans, have been shown to utilize oviposition deterrent pheromones (Prokopy

et al. 1984). These chemicals are detectable by conspecific females and provide information that a site, although desirable for oviposition in other respects, is already exploited. Perhaps one of the best known systems of this type is that of the apple maggot fly, *Rhagoletis pomonella* (Walsh) (Prokopy 1981b). Females of this tephritid oviposit into individual fruits, then drag the extended ovipositor across the fruit surface, depositing a pheromone which deters repeated egg-laying. Oviposition deterrent pheromones may also be associated with feeding larvae (Hilker 1985, Renwick and Radke 1981, Corbet 1971).

The following study tests the hypothesis that *C. rosaceana* uses some form of oviposition pheromone to regulate the spatial dispersion of egg masses. The first experiment examines the actual dispersion of egg masses under a normal laboratory rearing procedure. A second set of 3 experiments investigates the mechanisms behind this pattern in greater detail.

2.0 MATERIALS AND METHODS

2.1 EXPERIMENTAL PROCEDURES

All insects used in these experiments were newly eclosed *C. rosaceana* adults from the laboratory colony previously described. The temperature, R.H., and lighting were similar for the rearing and experimental rooms.

The first step in this study was to determine whether female moths altered their oviposition pattern in any way in the presence of conspecific egg masses. Such a mechanism would imply that the females were able to detect egg masses and, if confirmed, would justify further investigation. This objective was accomplished by conducting a dispersion analysis of egg masses on the 1100 cm^2 inner surface of the paper bags in which insects in the laboratory colony were allowed to eclose, mate, and oviposit. When ≥10 egg masses had accumulated in one of these bags, the remaining moths and pupae were removed, and the bag was opened out. Half of the egg masses were selected by random number, and the distance between each egg mass and its nearest neighbour was measured. Data were recorded for 20 such bags.

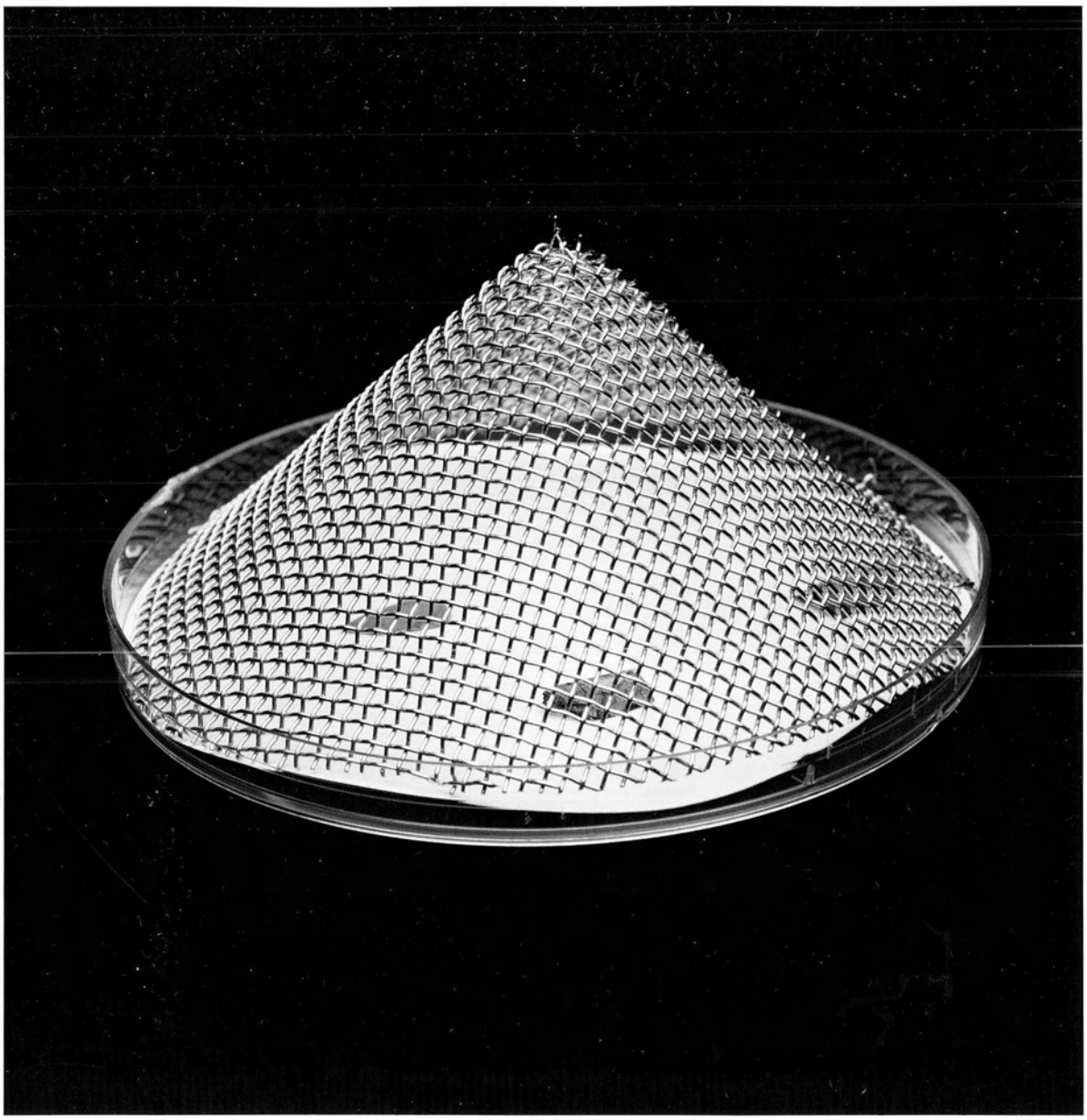
Since this preliminary study indicated that *C. rosaceana* females oviposit in a non-random, uniform dispersion pattern, a series of 3 experiments was conducted to investigate their response in greater detail. Oviposition chambers were

constructed, each made up of the inverted lid of a petri dish (9 cm diameter) covered with a wire mesh cone (Fig. 5). Moths would not oviposit on the mesh, but only on the floor of the chamber, which was covered with either filter paper or waxed paper. The moths were still able to fly, although in a very confined area.

One male and one female moth which had eclosed during the past 24 h were placed inside each oviposition chamber. The location of each female's first egg mass was recorded on the petri dish lid. The mated pair was then removed from the chamber and replaced with a second, newly-eclosed pair. The location of the second female's egg mass was also recorded on the outside of the petri dish lid. Observations were made between 1200 and 1300 h daily.

In the first experiment, filter paper was used as a substrate for oviposition. The initial egg mass remained attached to the filter paper when the second pair was introduced to the chamber. In the next experiment, filter paper was also used as a substrate, but the first egg mass was cut out of the paper and placed at a fixed location halfway between the centre and the perimeter at the front of the chamber. This experiment provided a control for any directional bias of oviposition in the first experiment, e.g. from an uneven distribution of light intensity. It also demonstrated the effect of handling the first egg mass. In the third experiment, waxed paper was used as an oviposition substrate, enabling the initial egg mass to be removed completely without damage. This procedure eliminated the

Fig. 5. Oviposition chamber used in study of *C. rosaceana* response to conspecific egg mass.



possibility of a visual or a tactile response to the rough surface of the first egg mass, and thus restricted any residual stimuli from the first egg mass to those of a chemical (pheromonal) nature. Each of the 3 experiments was replicated 30 times.

2.2 STATISTICAL ANALYSES

The dispersion analysis of egg masses in paper bags was based on a nearest-neighbour procedure developed by Clark and Evans (1954) for stationary objects. An index of dispersion, R, is calculated using a comparison of the actual mean distance to the nearest neighbour and the expected mean distance if egg masses were deposited randomly. If $R < 1$, the dispersion pattern is contagious, while if $R > 1$ the dispersion pattern is uniform. The standard error of this index can also be calculated, allowing the use of the standard normal statistic, Z. The probability value associated with Z is the level of significance of the difference between R and one.

The data collected from the oviposition chamber experiments were analyzed in two ways. For each experiment, a mean distance between first and second egg masses was calculated, along with its standard error. As well, radii were drawn from the centre of the chamber through the approximate centre of each egg mass, and to the fixed point at the "front" of the chamber. The resultant angles were analyzed using directional, or circular, statistics

as described below.

The radius drawn from the centre of the chamber through the egg mass, or through the front point is treated as a vector of magnitude 1. A sample of these vectors can be described and analyzed in several ways (Mardia 1972; Batschelet 1981). Phi-bar ($\bar{\phi}$) represents the mean angle of the sample, and is calculated as follows:

n = sample size

ϕ = angle formed by two vectors, i.e. between an egg mass and front or between second and first egg masses

$$\bar{x} = 1/n \, (\cos \phi_1 + \cos \phi_2 + \ldots + \cos \phi_n)$$

$$\bar{y} = 1/n \, (\sin \phi_1 + \sin \phi_2 + \ldots + \sin \phi_n)$$

if $\bar{x}>0$ *then*, $\bar{\phi} = \arctan (\bar{y}/\bar{x})$

if $\bar{x}<0$ *then*, $\bar{\phi} = 180^\circ + \arctan (\bar{y}/\bar{x})$

Confidence limits for $\bar{\phi}$ can also be determined (Mardia 1972). As well, it is possible to test whether a preferred direction is evident, using the non-parametric Rayleigh test (Batschelet 1981). This test requires calculation of the length of the mean vector, r, as follows:

$$r = (\bar{x}^2 + \bar{y}^2)^{0.5}$$

Ranges of r extend from 0 to 1.0, with uniform distributions of directions producing small values, and contagious distributions producing values close to 1.0, providing evidence of a preferred direction. Two assumptions are inherent in the Rayleigh test: 1) the data must have a unimodal distribution, and 2) individual observations must be independent (Judd and Borden 1988).

3.0 RESULTS AND DISCUSSION

3.1 EGG MASS DISPERSION PATTERN

All of the 20 paper bags examined had dispersion indices greater than 1.00 (Table 3). This pattern was significantly different from random, $P<0.05$, in 16 of the bags. Thus, *C. rosaceana* females do not oviposit randomly with respect to conspecific egg masses, but prefer to locate their eggs some distance away. Implicit in this result is that the females are capable of perceiving that a site is already occupied by developing eggs. In nature, these eggs would hatch before her own, and the resulting larvae would have a strong potential competitive advantage. A female could, therefore, increase her reproductive success by locating her eggs elsewhere (Prokopy 1981a).

3.2 INDIVIDUAL RESPONSE

Similar results were obtained for all 3 experiments conducted in the oviposition chambers (Table 4). Average distances ($\bar{x} \pm$ S.E.) between first and second egg masses were 3.98 ± 0.38, 3.95 ± 0.41, and 3.89 ± 0.34 respectively for the three experiments.

Using the front of the chamber as 0°, there was no evidence of a preferred direction for either the first (control) or

Table 3. Summary of results of Clark-Evans procedure applied to *C. rosaceana* egg masses oviposited in 1100 cm^2 paper bags.

Replicate	No. of egg masses	Index of dispersion (R)	Probability[a]
1	10	1.39	0.049*
2	17	1.49	0.007*
3	23	1.29	0.034*
4	10	1.52	0.014*
5	24	1.19	0.108
6	10	1.75	0.001*
7	10	1.60	0.006*
8	14	1.32	0.048*
9	17	1.59	0.001*
10	21	1.30	0.032*
11	15	1.31	0.046*
12	12	1.40	0.029*
13	14	1.28	0.078
14	18	1.26	0.067
15	10	1.59	0.006*
16	21	1.39	0.009*
17	14	1.57	0.002*
18	27	1.22	0.063
19	10	1.35	0.031*
20	12	1.39	0.030*

[a] Probabilities followed by asterisk indicate significantly non-random, uniform dispersion pattern.

Table 4. Angles (ϕ) of *C. rosaceana* egg masses from front of oviposition chamber (for control insects) or from location of previously oviposited egg mass (for experimental insects). Number of egg masses=30 in each case. Significance level of r (P) calculated from Rayleigh test. Experiments are as follows: A) filter paper substrate, first egg mass not disturbed, B) filter paper substrate, first egg mass relocated to front of chamber, C) waxed paper substrate, first egg mass removed.

	Control			Experimental		
Expt.	$\bar{\phi}$ (with 95% confidence limits)	Mean direction vector (r)	P of r	$\bar{\phi}$ (with 95% confidence limits)	Mean direction vector (r)	P of r
A	299.3 (±>90)	0.16	0.468	185.5 (±26)	0.55	<0.001
B	283.1 (±>90)	0.03	>0.900	199.2 (±32)	0.44	0.002
C	299.2 (±>90)	0.07	0.864	173.3 (±25)	0.54	<0.001

second (experimental) egg masses. However, rotating the location of the first egg mass to 0° for analysis resulted in a significant preferred direction for the second egg mass (Table 4). Since the mean angle for experimental insects was not significantly different from 180° (Table 4), it can be concluded that these moths were exhibiting an avoidance response to the first egg mass. Because these results held for the second experiment, neither the orientation of the chamber nor the handling of the first egg mass had any effect on the moths' response. More importantly, the modification of oviposition pattern persisted in the third experiment, even though the first egg mass was no longer present. Therefore, the mechanism for egg mass detection is chemically mediated, rather than tactile or visual. An epideictic or spacing pheromone (Prokopy 1981a) was evidently left impregnated in the wax surface of the arena.

From observations of *C. rosaceana* oviposition behaviour, it can be hypothesized that the epideictic pheromone is associated with scales on the ventral surface of the female's abdomen. Such a phenomenon has been proposed by Prokopy (1981a) for lepidopterans in general. When ovipositing, a female moth deposits eggs on alternate sides of the growing egg mass. As a result, she drags her abdomen across the hardening eggs repeatedly. Numerous scales can be observed embedded in the egg mass. Kairomones emanating from such scales are used in host selection by several predators and parasitoids (Lewis *et al.* 1977, Prokopy 1981a). The same semiochemicals may also be used

as epideictic pheromones. Examples of this phenomenon include the aggregation pheromones of bark beetles which are used as host-finding kairomones by entomophagous insects (Borden 1982) and the mandibular gland secretions of *Ephestia kuehniella* which contain both an epideictic pheromone and a kairomone for parasitoids (Corbet 1971, Corbet 1973). It is also possible that the pheromone is deposited with the egg mass from the ovipositor, possibly in the cement, and is chemically related to, or includes one or more components of the sex pheromone (Vakenti *et al.* 1988). Such multiple functions would be in accordance with Blum's (1970) theory of "pheromonal parsimony".

CONCLUSIONS

My results show clearly that *C. rosaceana* is adversely affected by high larval densities. The effects of crowding on the life history of this insect include lower survival, smaller pupal size, more time spent in the pupal stage, and an altered larval dispersal pattern. All of these factors could combine to lower individual fitness as measured by reproductive success (Prokopy 1981a). As a result, it was hypothesized that one or more mechanisms exist which enable the insect to avoid larval crowding. The examination of oviposition patterns upholds this hypothesis and indicates that an epideictic pheromone is associated with recently-laid egg masses. In the wild, such a pheromone would enable individual moths to increase their reproductive fitness by avoiding occupied sites, thus minimizing competition for their offspring.

Density-dependent responses, such as those exhibited by *C. rosaceana*, can have numerous practical implications. The use of pheromones to induce pest insects to disperse out of an area is a relatively new concept, but it has been used successfully in some cases (Kline *et al.* 1974, Katsoyannos and Boller 1980, Lindgren *et al.* 1989). However, it is unlikely that epideictic pheromones alone will be able to control any pest problem (Prokopy 1981a). Thresholds may change over time, such that in the absence of untreated sites oviposition or feeding may occur at treated sites. The disadvantages of intraspecific competition will never outweigh the disadvantages of starvation, or dying without ovipositing. Density-dependent responses may be best

exploited in the context of an integrated management program. For example, as proposed for bark beetles, epideictic pheromones could be used to control the density of an infestation, so that host resistance mechanisms remain effective (Hedden and Pitman 1978). They may also have potential as stressing agents (Jaques 1962), enhancing population management by increasing the effectiveness of chemical or microbial pesticides.

Much more work is required in this field. Several other density-regulating systems could be operative in *C. rosaceana*. The behavioural mechanisms of density-dependent responses must be investigated in greater depth, and pheromonal components of the systems should be isolated and identified. The search for epideictic pheromones should be extended to other pest insects. As well, thresholds need to be established, both for the density responses themselves and for the tolerance level of the individual to an epideictic pheromone.

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