

AN ABSTRACT OF THE THESIS OF

Santiago Martí Martí for the degree of Doctor of Philosophy in Entomology presented on July 12 2000. Title: Host Selection, Reproductive Biology, Host-specific Development and Mortality of the Codling Moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), in Apple and Pear.

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Abstract approved: _____

Helmut Riedl

The biology and behavior of the codling moth, *Cydia pomonella* (L.), was studied to determine factors responsible for differences in susceptibility to moth damage among apple and pear cultivars.

In release experiments, codling moth adults were intercepted in significantly greater numbers in host (apple, pear) than in non-host (maple) trees, suggesting a directed response rather than random movement to host trees. There was no correlation between adult preference for certain hosts and the susceptibility of the hosts to codling moth attack. Adults were guided by cues from the tree canopy, foliage and fruit. Cues from host fruit were less important at the beginning of the season. Similar adult behavior patterns were observed in different codling moth strains. However, moths reared on apples were guided more by olfactory cues from fruit than sterilized moths reared on artificial diet.

The percentage of moths mating under caged conditions was higher in apple and pear than in maple trees. In host plants, stimuli from fruit were not essential for mating. The egg distribution in the field varied through the season depending on the host cultivar. However, mean distance of eggs to fruit, as well as larval travel speed, was not different among host cultivars. Selection of an oviposition site by the adult female was affected by several factors, including visual, chemical, and tactile stimuli of host fruit, as well as anatomical (pubescence) and chemical (oviposition stimulants) properties of leaves.

There were no differences in natural mortality and developmental rates of the egg stage on host cultivars. However, there were significant differences in first instar larval mortality among hosts over most of the season. Mortality was much higher on 'Anjou' than on 'Bartlett' and 'Red Delicious', except at the end of the season. Differential first instar mortality was due to the different rate of success in entering the fruit. The ability to penetrate fruit was correlated with infestation levels observed in the field and related both to neonate behavior and anatomical characteristics of host fruits.

Larval food source did not affect larval and pupal development rates, adult fecundity, or egg viability.

**Host Selection, Reproductive Biology,
Host-specific Development and Mortality of the Codling Moth,
Cydia pomonella (L.) (Lepidoptera: Tortricidae), in Apple and Pear**

by

Santiago Martí Martí

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**Host Selection, Reproductive Biology,
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Cydia pomonella (L.) (Lepidoptera: Tortricidae), in Apple and Pear**

CHAPTER 1

INTRODUCTION

The codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), is an important worldwide pest of pome fruits, some stone fruits, and walnuts. Apples and pears are its preferred hosts. There are many faunal and horticultural distinctions between these two tree fruits which are pertinent to the management of their respective pest complexes. An understanding of the relationships between a pest and its hosts is important for optimal management. This is also true for codling moth. Many aspects of codling moth biology, ecology, behavior, and control have been intensively studied on apples, its most widely distributed and important host. However, the scope of codling moth research on other hosts including pears has been limited and considerable gaps in understanding of codling moth biology and behavior remain.

In the Hood River valley of northern Oregon, apples and pears are often planted in close proximity to each other and they are the principal hosts of codling moth in this area. Apples are considerably more susceptible to codling moth injury than pears. Differences in susceptibility seem to be more pronounced among pear than among apple cultivars. On apple, differences in infestation levels among early and late cultivars have been related to the number of codling moth generations a cultivar is exposed to in the field. In addition, certain intrinsic fruit properties such as large fruit size, unusual amounts of fragrance, tender skin, and degree of sweetness, have been related to higher susceptibility to codling moth injury. Among the pear cultivars, 'Bartlett' has been shown to be more susceptible to larval entry than any of the winter pear cultivars such as 'Anjou' and 'Bosc'. The resistance to codling moth in winter pears has been related to certain anatomical characteristics (e.g., stone cells) of the fruit which act as a physical barrier to larval entry. It has also been suggested that the

presence of certain fruit odors which may affect adult reproductive and larval behavior might be partly responsible for the observed differences in susceptibility to codling moth.

Differences in codling moth infestation levels may be a manifestation of host preference by the adult stage, differential mating and oviposition activity and/or differential mortality of immature codling moth stages on different hosts. In spite of more than 100 years of codling moth research, the literature offers little information about questions related to host finding and host selection in the codling moth. The questions of how codling moth locates hosts is particularly relevant in light of the increasing use of mating disruption (MD) for codling moth control where immigrating moths can easily compromise the success of the MD-based control program. Factors affecting oviposition by mated females need to be studied in more depth to provide information about egg distribution in different host cultivars. Egg distribution on a host is indirectly related to susceptibility since neonate mortality can be expected to be higher in those cultivars where eggs are laid farther away from the fruit. In addition, cultivar-specific resistant factors associated to low susceptibility to codling moth larval penetration need to be identified. Food quality may vary with the type of fruit or medium on which larvae feed and it may affect not only the developmental rate but also fecundity of the adult and egg viability. Therefore, the reproductive potential of codling moth populations on different hosts may not be the same which could contribute to differences in infestation levels in the field.

The objectives of the research presented here are to investigate several key aspects of the relationship between the codling moth on its principal hosts and to determine the factors responsible for differences in codling moth susceptibility among apple and pear cultivars. The aspects of the biology and behavior of the codling moth studied include (1) host selection by the adult stage, (2) mating and oviposition site selection by the adult female, (3) egg distribution in the field on different apple and pear cultivars, (4) searching behavior of neonate larvae to locate and penetrate the fruit, (5) host-specific mortality factors affecting immature stages, and (6) the relationship between larval food source, post-embryonic development and reproduction. A better understanding of the factors responsible for the differences in codling moth susceptibility could be used to develop cultivar-specific control programs, thereby reducing the need for chemical intervention.

CHAPTER 2

LITERATURE REVIEW

2.1 Biology and distribution

The codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), is a widely distributed pest of cultivated pome fruits, some stone fruits, and walnuts. Depending on climate and food availability the codling moth has one to four generations in its range of distribution (Shel'Deshova 1967, Riedl 1983).

Codling moth has four life stages: egg, larva (5 instars), pupa, and adult. The codling moth overwinters always as mature diapausing larvae in a cocoon on the trunk or main limbs of the host tree, or in protected places such as litter on the ground, pruning wounds, and bin storage areas. In the spring, pupae are formed from overwintering larvae. Adults emerge from the pupae, and females lay their eggs on the foliage, fruit, or wood of the host tree. Damage occurs during the larval stage. Larvae feed both on the flesh and seeds of fruit. Mature, fifth-instar larvae, leave the fruit in order to search for a cocooning site. These larvae either pupate and emerge as the next adult generation or enter diapause.

Diapause is the principal mechanism for coordinating the codling moth life cycle with the variation of temperature and food conditions (Shel'Deshova 1967). In temperate zones, diapause is induced by changes in the photoperiod. Other variables, such as temperature, food availability and food quality, can play a role, especially in areas where the photoperiod fluctuates little through the year (i.e., near the equator). Diapause in the codling moth is facultative, although a certain proportion of larvae, that display no photoperiodic response, is genetically univoltine (Riedl 1983).

The codling moth occurs wherever apples and pears are grown, except for the Korean Peninsula (Park & Park 1976), Japan and the mainland of Asia east of about the 90th degree of longitude (Chapman & Lienk 1971), and Western Australia where it has been eradicated (Barnes 1991). Its origin is Eurasian. The distribution of the codling moth is affected especially by ecological factors. The northern boundary is defined by temperature, i.e.

adequate heat summation above 10°C in summer of approximately 600 degree-days (° C), the heat units required for development of one generation (Riedl & Croft 1978). In both northern and southern hemispheres, its limits of distribution approaching the equator are approximately at the 25th parallels (Shel'Deshova 1967).

2.2 Economic importance and control

In most of the deciduous fruit-growing areas around the world, codling moth is the key pest of pome fruits and its control dominates the control practices for the whole pest complex (Riedl et al. 1998). Codling moth damage must be kept low since the market has essentially a zero tolerance for infested fruit. This is the reason why insecticides have been the primarily used tactic for codling moth control for many years. Approximately half of the costs in controlling arthropod pests of apples in the state of Washington are attributable to the control of the codling moth (Westigard et al. 1976).

After resistance developed to inorganic insecticides, such as lead arsenate (Hough 1928), DDT (Cutright 1954), then organophosphates (azinphosmethyl), carbamates (carbaryl), and more recently pyrethroids have been used against codling moth (Croft & Riedl 1991). Organophosphates have been the principal tools for codling moth control for the last 50 years, but the development of resistance has compromised their use for codling moth control in many areas, including California (Welter et al. 1992), Washington (Croft & Riedl 1991), South-Africa (Blomefield 1994), and Europe (Sauphanor et al. 1996). To avoid resistance problems in the future, codling moth control will have to rely on the combination of several different control tactics with different modes of action (Riedl et al. 1998). Resistance-prone control tactics, such as broad-spectrum insecticides and insect growth regulators (IGRs), should be alternated with control tactics with low resistance risk (e.g., microbial insecticides, botanical insecticides, oil sprays, mating disruption, biological control, plant resistance, cultural controls, etc.).

Some insect growth regulators, including chitin-synthesis inhibitors (e.g., diblubenzuron), juvenile hormone mimics (e.g., fenoxycarb), and ecdysone agonists (e.g.,

tebufenozide), are registered for codling moth control in some crops in Europe, Argentina, South Africa, and the United States (Riedl et al. 1998). However, both in Italy (Riedl & Zelger 1994, Sauphanor et al. 1997) and France (Sauphanor et al. 1994) populations resistant to diflubenzuron have been detected.

Currently botanical insecticides such as ryania, produced from a tropical woody plant, are used to a very limited extent (Riedl et al. 1998). One botanical insecticide which may have some promise for codling moth control is azadirachtin. This is a natural constituent of neem oil which is extracted from neem (*Azadirachta indica*) seeds. Azadirachtin acts as an antagonist of the molting hormone ecdysone (Riedl et al. 1998).

Avermectins are fermentation products isolated from *Streptomyces avermitilis* Burg. This group of insecticides may be helpful for codling moth control mostly in early season when young larvae are generally less successful at entering fruit (Croft & Riedl 1991). Spinosad, produced by another soil microbe *Saccharopolyspora spinosa*, may be helpful for codling moth control in conjunction with other control tactics (Riedl et al. 1998). Other microbial insecticides, such as granulosis virus ('CpGV'), and *Bacillus thuringiensis* have the problem of short persistence and are easily degraded by sunlight. In addition, they do not act by contact and larvae must ingest a toxic dose before entering the fruit, which requires good spray coverage. Entomopathogenic nematodes, such as *Neoplectana carpocapsae* Weiser and *Steinernema carpocapsae* (Weiser), have been used in research trials to control cocooning larvae in fruit bins (Lacey & Chauvin 1999).

The use of oil sprays during the growing season to assist with control of codling moth is promising because of their selectivity to beneficials and safety to the applicator and consumer. The major concern with oil use after bloom is the potential phytotoxicity to fruit and foliage as well as long-term negative impact on trees (Riedl et al. 1998).

The use of mating disruption, as well as other control tactics that use pheromones, is very promising for codling moth control, but it is not a suitable control method for every orchard situation. Orchard size, shape, and topography need to be considered for successful use of this control tactic (Riedl et al. 1998). Blomefield (1995) considered codling moth a good candidate for mating disruption mostly because its sexual communication is primarily olfactory and females usually mate only once and have limited range of migration if fruit is

present. Currently the complementary use of mating disruption and two or three applications of organophosphates is a widely used tactic of control in the Pacific Northwest.

The codling moth has many natural enemies including birds, spiders, predaceous and parasitic insects, nematodes, bacteria, fungi, protozoa, and viruses (Falcon & Huber 1991). However, codling moth control in commercial orchards cannot rely on biological control. Parasitism due to the egg parasitoid, *Trichogramma minutum* Riley (Hymenoptera: Trichogrammatidae), and the egg-larval parasitoid *Ascogaster quadridentata* Wesmaels (Hymenoptera: Braconidae) may be important, but generally they cannot control codling moth at commercially acceptable levels (McLellan 1962). Some bird predators, mostly woodpeckers, have also been shown to be efficient biological control agents in some cases (MacLellan 1962).

Horticultural practices such as irrigation, pruning and fruit thinning, may also impact the codling moth. The choice of rootstocks and tree training may affect the success of the codling moth by limiting the number of cocooning sites on the tree (Blomefield 1991, Riedl et al. 1998). Since first-instar larvae find it easier to enter where fruit contact each other, thinning practices leaving single fruits in a cluster may help with codling moth control. This practice will make it more difficult for young larvae to penetrate the fruit and allow better spray coverage of the fruit surface (Riedl et al. 1998). The use of overtree irrigation may reduce codling moth damage by up to 60-90%, but may increase disease pressure (Knight 1998, Riedl et al. 1998).

2.3 Hosts

The codling moth is an oligophagous species that attacks a variety of tree crops ranging from pome and stone crops to nut crops. All principal hosts, such as apple, pear, quince, apricot, and plum, are members of the Rosaceae family, except for walnut (Juglandaceae). Although codling moth has been reported as a pest of some importance on apricots in California and South Africa (Petty 1926, Madsen & Borden 1954), and the Persian walnut, *Juglans regia*, in California and Europe (Quayle 1926, Putman 1963), it is most destructive to apple and pear. It can develop in cherries (Putman, 1962) and in

peaches (Petty 1926, Bovey 1949, Madsen & Borden 1954). Other incidental hosts are the chestnut, *Castanea* spp. (Fagaceae), persimmon, *Diospyros* spp. (Ebenaceae), hawthorn, *Crateagus* spp. (Rosaceae) and ornamental crab-apple, *Malus* spp. (Rosaceae) (Putman 1963).

Several early studies have been conducted to determine differences in susceptibility to codling moth attack, mostly on apple cultivars, and determine the factors responsible for differences in susceptibility. Felt (1910) observed a higher percentage of infestation on 'McIntosh' trees than on 'Wealthy'. Newcomer & Yothers (1924) noticed that cultivars such as 'Winesap' and 'Arkansas' were less injured than some of the more fragrant cultivars, such as 'Spitzenburg' and 'Delicious'. Cutright & Morrison (1935), and Witehead (1944) observed that codling moth damage was relatively low on 'Jonathan'. Most of these authors related low susceptibility to early ripening and harvest prior to emergence of the later codling moth generations. Notwithstanding, Cutright & Morrison (1935) and Witehead (1944) indicated that some cultivars (e.g., 'Jonathan', 'Winesap') also possessed some resistance factors other than escape in time due to early maturity. Fruit characteristics as above average fruit size ('Wolf River', 'Fallwater'), unusual amounts of fragrance ('Delicious', 'Spitzenburg'), tender skin ('Chenango', 'McIntosh'), and subacidness or sweetness ('Paradise Sweet', 'Delicious') were associated with higher susceptibility to codling moth injury.

Cultivar susceptibility to codling moth injury on pear cultivars has been studied to a limited extent. Westigard et al. (1976) observed that 'Golden Delicious' apples were more susceptible to codling moth than any pear cultivar. Among the pear cultivars studied, 'Bartlett' pears were more susceptible to larval entry than any of the winter pear cultivars such as 'Anjou' and 'Bosc'. He related resistance to codling moth penetration on winter pears to the presence of a layer of stone cells. Later studies (Russ 1976, Sutherland et al. 1977) suggested that cultivar differences in fruit odors both on apples and pears, including certain specific constituents that might act as mating and/or oviposition stimulants and/or larval attractants, could be related to cultivar susceptibility.

2.4 Flight, dispersal, and host selection

Codling moth larvae can not choose among potential host trees because they travel only very short distances and can survive for only a short time without feeding. Consequently, codling moth host selection is restricted to the winged adult stage which may display a preference for a host tree over a non-host tree, for apple over pear or for a specific cultivar when given a choice.

Environmental factors affecting codling moth flight have been extensively studied. This information has been useful for interpreting catches in pheromone traps used for many years to monitor codling moth populations and make management decisions (Riedl et al. 1986). Codling moth flight may be described as a quick, zig-zag, movement at various angles and may rise or drop suddenly. Both sexes have similar flight habits (Worthley 1932). Borden (1931) reported that codling moth was not able to sustain long flights (only 20 to 25 m). This idea has been rejected by other authors (Steiner 1940, Geier 1963, White et al. 1973, Suski et al. 1981) who observed flights longer than 800 m.

Flight is governed by a complex of environmental factors, including light intensity, temperature, humidity, and air movements. Borden (1931), using bait traps, and Zech (1955), using UV-light traps, determined that the daily flight peak is around sunset. Wong et al. (1971) determined that the daily rhythm of male response to virgin females was also around sunset. Batiste et al. (1973) and Riedl & Croft (1975) observed the same periodicity with synthetic pheromones. Environmental factors such as rain and temperatures prior to sunset can shift the daily activity rhythm. Cool temperatures will advance it, but warm afternoon temperatures will delay it (Batiste et al. 1973). Zech (1955) observed an increase in flight activity just before the onset of rain. The duration of codling moth flight has been reported to extend over a period of approximately two (Borden 1931) to five hours (Batiste et al. 1973) under favorable conditions. Chernyshev (1961) attributed this daily flight rhythm not only to a reaction to daily changes in external conditions, but also to an internal rhythm. A second activity peak can occur following sunrise, but it is generally much smaller and less frequent than the evening flight (Borden 1931) because of low temperatures (Zech 1955, Pristavko 1971). Different temperatures have been reported as lower

thresholds for codling moth flight activity: 10 to 12° C (Bauckmann 1956, Pristavko 1969); 12 °C (Russ 1961, Klinger et al. 1958); 12 to 13 °C (Batiste et al. 1973); 12 to 14.5 °C (Borden 1931); 14 to 16 °C (Zech 1955), and 15.5 °C (Parrott & Collins 1934). Different temperature ranges have also been reported as the optimum for flight: between 15 and 26 °C (Pristavko & Chaika 1976), between 20 to 25 °C (Nikolic 1958, Lekic 1958). Batiste et al. (1973) reported a decrease in flight activity above 27 °C. Wind affects flight activity adversely, even at very low wind speeds, although there is no general agreement on the threshold that inhibits flight. Wind above 1 m/sec reduced UV-light trap catches and above 4 m/sec inhibited flight completely (Pristavko 1969). However Russ (1961) indicated that wind up to 5 m/sec had little effect on flight activity but was inhibitory at higher wind speeds. The effects of humidity and changes in humidity are not really known except that high humidity inhibits flight activity (Russ 1961). Rain also negatively affects flight activity, even at very low amounts. In one study, amounts above 0.2 mm/h almost completely inhibited flight (Russ 1961). Worthley (1932) mentioned that flight was prolonged by moonlight and was favored by high barometric pressure.

Little is known about how the adult codling moth finds a host tree, whether the search is random at first or whether long-range attraction is involved. Insects perceive and integrate stimuli detected by olfactory, visual, tactile, and gustatory receptors. Olfactory and visual stimuli may play a key role in directing insects to a host plant. Host plants may emit attractants and/or arrestment odors, while non-host plants may lack attractants and/or arrestment odors or may emit repellent odors. Visual cues received by insects while searching for a host plant may include the spectral quality of the light stimuli, as well as the dimensions and shape of the objects viewed (Schoonhoven 1968, Smith 1989). During long-range orientation, insects perceive and integrate simultaneously visual and olfactory stimuli and they can use vision for recognition of the shape, color or size of an object and olfaction to perceive plant attractants. After approaching the immediate location of the plant, movement to the host plant may also be due to the perception of flight arrestment odors and the outline of the plant (Schoonhoven 1968, Smith 1989). Few studies have focused on the potential role of olfactory stimuli directing adult codling moth to the host plant. Wearing et al. (1973) showed that exposure to apple odor resulted in an increase in locomotor activity

of mated females. Buda (1978) determined that the odor of extracts from young apple leaves and flowers raised the level of locomotor activity of codling moth males. Skirkevicius et al. (1980), using electroantennogram studies, also demonstrated that codling males are excited by odors emanating both from apple fruits and leaves. Skirkevicius & Tatjanskaite (1971), working with the chemical attractant geraniol, determined that the sensitivity of the codling moth to olfactory stimuli depends also on the time of day.

Wildbolz (1958) studied codling moth dispersal in a uniform apple orchard. He concluded that movement of codling moth females between closely planted trees was random as a result of the much greater aggregation of eggs observed in the immediate vicinity of the liberation point than would be predicted by a normal cumulative distribution. The spatial distribution of offspring fitted a Bessel function, which suggests random movement of moths in uniform surroundings. He considered that female movement from tree to tree in an orchard was guided mostly by visual stimuli, and randomness in flight direction and length depended upon uniformity in the distribution of potential oviposition sites. He hypothesized that a continuous succession of oviposition sites within easy flying distance would result in maximum dispersion of oviposition females around a source. The mean distance traveled in single flights would increase at longer distances between potential hosts. At longer distances between potential hosts the probability of successful host selection would decrease and the probability of moth death would increase. In another dispersal study, Suski et al. (1981) hypothesized that lower recaptures of male moths within an apple orchard were associated with lower fruit set due to frost injury at blossom time.

2.5 Mating behavior

There is limited information about the effect of stimuli from host plants that trigger mating on female codling moths. Castrovillo & Cardé (1979) determined that, like the other reproductive behaviors in the codling moth, mating followed a circadian rhythmicity taking place during late evening or after sunset, depending on temperature. A decrease in temperature lead to a reduction in the proportion of females calling when the decrease occurred during the scotophase and a shift in peak calling into the photophase when the

decrease in temperature occurred some hours before the initiation of the scotophase. Castrovillo & Cardé (1980) also observed that chemical stimuli provided by synthetic pheromone guided male codling moths to the source of sex pheromone over long distances; at short distances visual cues from female moths seemed to be more important. Hutt & White (1977) also showed that visual stimuli from female increased male codling moth copulatory activity. In the field, especially in the spring, mating may be delayed and egg laying may not take place for several days or even a week or two after mating when temperatures are low. Deseö (1970) indicated that virgin females already contain mature oocytes immediately after emergence. Gehring & Madsen (1963), Nowosielski & Suski (1977) and Howell et al. (1978) observed in laboratory studies that codling moth virgin females were ready to mate and lay eggs within 24 h after emergence. Maximum mating activity occurred between the second day (Nowosielski & Suski 1977) and the fourth day of adult life (Gehring & Madsen 1963). Howell et al. (1978) also showed that mating was influenced both by the ratio of males to females and the density of moths present. A reduction in the ability of females to attract males after mating was observed by Gehring & Madsen (1963). White et al. (1975) and Howell et al. (1978) determined that copulation of virgin moths usually lasted about 40-50 minutes; subsequent matings were longer and the size of the spermatophore was reduced. Males usually need more than 12 hours after mating to produce a second spermatophore (Howell et al. 1978).

2.6 Oviposition behavior

For the codling moth, the selection of a specific plant part as a suitable oviposition site in close proximity to the larval food source is crucial because of the mobility limitations of neonate larvae. Although the adult codling may oviposit in absence of fruit or any food suitable for larval development (Deseö 1970), female moths probably locate suitable oviposition sites on its host plant by following certain stimuli. Visual, tactile, and chemical (olfactory) stimuli may be involved (Deseö 1970).

Many authors have observed the avoidance of female moths to oviposit on pubescent surfaces on apple (Hall 1929, VanLeeuwen 1939, MacLellan 1962, Putman 1963, Geier 1963, Wood 1965, Hagley et al. 1980), walnut (Olson 1977) and peach (Yokoyama & Miller 1988a, Curtis et al. 1990). Similarly, the oriental fruit moth, *Grapholita molesta* (Busck), avoids the heavily pubescent surface of peach fruits (Yokoyama & Miller 1988b). Plourde et al. (1985) related codling moth egg distribution to fruit penetration and infestation levels and suggested that apple cultivars having a more pubescent lower leaf surface had significantly lower fruit damage especially in the first generation. Balensky et al. (1973) reported egg deposition by codling moth females on downy young almond fruit. Hagley et al. (1980) suggested that the density of leaf hairs was not the only physical factor affecting egg distribution. The nature of the wax scales on some surfaces (i.e. upper side of pear leaves) also appeared to influence the number of eggs deposited.

Several authors reported an effect of naturally occurring compounds and host odors on codling moth oviposition. Dethier (1947) concluded that essential oils such as citronellol, oil of cloves, and pine tar oil act as codling moth oviposition stimulants. VanLeeuwen (1947) suggested the use of apple odor in the laboratory as an oviposition stimulant. In the field, gravid codling moths were stimulated to oviposit by the odor of apple fruits (Wildbolz 1958). Wearing et al. (1973), in laboratory studies using an olfactometer, confirmed that moths lay more eggs when an apple source was present. This could involve physical/contact and/or chemical stimuli. They also found that apple odor not only stimulated oviposition but also affected egg distribution. When an apple odor was present, a higher proportion of eggs was laid close to the odor source where the air stream entered the olfactometer flight chamber. This is consistent with the findings of several authors concerning the distance most eggs are laid from the fruit. Wearing & Hutchins (1973), Wearing et al. (1973), and Sutherland et al. (1974) suggested that alpha-farnesene, a volatile sesquiterpene released by apple (Murray et al. 1964, Huelin & Murray 1966, Meigh & Filmer 1967), pear (Jennings & Tressl 1974) and quince fruits (Shimizu & Yoshihara 1977), may act as an attractant to female codling moths. (E,E)- and (Z,E)- α -farnesene, the two isomers which occur naturally (Anet 1970) have been implicated as oviposition stimulants for gravid female moths and attractants for newly hatched codling moth larvae (Sutherland 1972, Sutherland & Hutchins

1972, 1973, Wearing & Hutchins 1973, Sutherland et al. 1974, Gut 1980). Russ (1976), Sutherland et al. (1977), and Gut (1980) suggested that cultivar differences in fruit odors, including certain specific constituents such as α -farnesene that might act as mating and/or oviposition stimulants and/or larval attractants, could be related to cultivar susceptibility. Russ (1976) indicated that gravid moths were more attracted to apples than pears. He suggested that both cultivar preference and fruit attractiveness change through the season and were correlated with changes of α -farnesene during fruit development. Sutherland et al. (1977) reported an increase of the total α -farnesene level in apple fruits during fruit growth and maturation. Gut (1980) found different results and reported that codling moths preferred pear to apple cultivars when given a choice between the two. Preference was observed late but not early in the season, indicating that it was a result of factors associated with the seasonal growth of the plants. He also found that the total amount of α -farnesene per fruit and per unit area of skin (range from 1 to 9 $\mu\text{g}/\text{cm}^2$) was greater in the pear cultivars studied than in apple cultivars, and the amount per unit area decreased from early to late season. In preference tests, he found a positive correlation between concentration of α -farnesene and oviposition on apple and pear cultivars. Oviposition behavior may be affected by more than one olfactory stimulant. Sutherland et al. (1974) suggested that normal oviposition depends upon the presence of more than one olfactory stimulant/arrestant and that some terpenoids, such as geraniol and citral, might play an important role. Both compounds were isolated from apples and had a positive influence on the sexual behavior of male codling moths. Host odors stimulate oviposition also in other phytophagous species, such as the bean weevil, *Acanthoscelides obtectus* Say, (Labeyrie 1960), the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), (Sanders 1962), the cabbage weevil, *Ceuthorrhynchus macula-alba* Hrbst (Saringer 1963), *Scrobipala ocellatella* Boyd. (Robert 1964), and the bean seed fly, *Chortophila brassicae* Bouché (Zohren 1968). Gut (1980) suggested that low oviposition on ripened fruit could be associated with volatiles, such as ethylene, that are released when fruit reaches maturity and that may act as oviposition deterrent. Thiéry et al. (1995) suggested that compounds present on codling moth egg extracts may affect oviposition site selection and lead to a dispersive oviposition behavior.

Deseö (1970) suggested that the beginning of oviposition and egg production in the codling moth do not seem to be influenced by the absence of the host fruit. Codling moth oviposition may be influenced not only by host stimuli but also by different environmental conditions, such as temperature, light intensity, rainfall, humidity, and air movements. Environmental conditions must be favorable for egg laying to occur (Gut 1980). Several authors (Siegler & Plank 1921, VanLeeuwen 1929, De Jong & Post 1963, Riedl & Loher 1980) observed that, similar to mating, oviposition follows a circadian rhythm. It takes place toward the end of the day with a distinct peak before sunset. Several authors (Isley & Ackerman 1923, Hall 1929, Cutright 1937, Isley 1938) observed that temperatures below 15.5-16.5 °C inhibit oviposition of caged moths. Parker (1959) reported a low threshold of 18.3 °C and Klinger et al. (1958) a threshold of 12°C. Oviposition by first and second generation codling moth females was highly correlated with the average temperature between 5-11 p.m. (Hagley 1976). Isley (1938) found that egg laying increased with temperature to about 27° C, but declined above this point. Hagley (1976) determined that the optimum temperature range for oviposition in the laboratory was 23-25 °C. Egg production is also influenced by the conditions the female moth was exposed to in early stages of development. Isley (1938) and Proverbs & Newton (1962) observed a reduction in egg production in moths exposed to high temperatures in the larval and pupal stages. Dickson et al. (1952) reported that the rate of oviposition was greater under natural light conditions as opposed to artificial light. The degree to which both flight and egg laying are suppressed by air movement has not been investigated in depth. Parker (1959) observed that oviposition by moths confined in a greenhouse was prevented by an air current of four miles per hour. Hagley (1976) reported that oviposition was also adversely affected by the amount and duration of rainfall occurring between 5 and 11 p.m.

2.7 Egg distribution

Several authors have studied the distribution of codling moth eggs, especially in apple trees. Female moths usually lay eggs singly on the fruit or on the leaves near the fruit, although egg-laying patterns have been shown to vary with host species and cultivar and with the seasonal development of the fruit. Physical and chemical factors have been proposed to explain the variation in the distribution of codling moth eggs. Studies on the distribution of eggs among fruit clusters of apple varieties have yielded contradictory results. Geier (1963) indicated that eggs were distributed randomly with regard to individual fruits and that the frequency of oviposition within clusters of one, two, and three fruits per cluster was also random. He concluded that each fruit had an equal instantaneous probability of being used as oviposition site. Wood (1965) suggested that the frequency distribution of eggs on individual fruit clusters was mildly contagious (partially due to weather conditions and not due to female behavior) and oviposition within clusters of one, two, and three or more fruits per cluster was random on 'Delicious' but not on 'Gravenstein'. On the other hand, Jackson (1979) indicated that the distribution of eggs among clusters deviated from randomness and he hypothesized that each fruit has a 'sphere of influence' around it, which affects the oviposition behavior of female moths. He considered that the 'spheres of influence' of fruits do not have an additive effect where they overlap. Hall (1929) indicated that early in the season eggs are mostly laid on the upper leaf surface on apple cultivars, in mid-season on the underside, and later after the pubescence wears off mostly on the fruit. VanLeeuwen (1939) also checked the within-cluster egg distribution on three apple cultivars. He found approximately 53 % of the eggs on the upper surface of primary leaves, 22 % on the upper surface of secondary leaves, 4 % on the lower surface of primary leaves, 2 % on the lower surface of secondary leaves, 15 % on the fruit, and 4 % on the twigs and stems. On 'Jonathan' and 'Winesap' trees practically all eggs were on the upper surface of the leaf, whereas on 'Rome Beauty' most were on the lower surface. Putman (1963) reported that from 80 to 95 % of the eggs laid on apple trees were on the leaves, mostly on the upper surface, 5-18 % on the fruit, and 0-2 % on the twigs. He also noted that the percentage of eggs laid on the fruit increased late in the season. Geier (1963) found that approximately one-quarter of the eggs laid on 'Granny Smith' and 'Delicious' apple trees

were on fruits, one-fifth on lower leaf surfaces and more than one-half on upper leaf surfaces. He also found the proportion of eggs on lower leaf surfaces to be relatively constant, whereas the proportion of eggs on fruit was as low as 14 % in the spring, rose to 40 % in mid-summer, and decreased gradually to 20 % towards autumn. Wood (1965) found some differences on egg distribution in 'Gravenstein', 'Delicious', and 'Sturmer' apple cultivars. Fruit and upper side of the leaf were the preferred oviposition substrates, but twice as many eggs were laid on the lower leaf surface of 'Delicious' than the other two cultivars. Jackson (1979) and Subinprasert & Svensson (1988) also observed a preference for the upper side over the underside of leaves and fruit on different apple cultivars. Blomefield et al. (1997) checked oviposition both in 'Granny Smith' and 'Golden Delicious'. The preferred oviposition sites on both apple cultivars were the leaves. Wood was the least preferred. On leaves, the upper surface was preferred at the beginning and end of the season and the lower surface during the middle of the season in both cultivars. More eggs were laid on the fruit of 'Granny Smith' (35.6 %) than on those of 'Golden Delicious' (10.7 %).

There are few references about the distribution of eggs on pear cultivars. In South Africa, Hattingh (1942) sampled three pear cultivars ('Beurre Bosc', 'Bon Chretien', and 'Winter Nelis') for oviposition and found that spring brood moths deposited most of the eggs at full bloom. As the season advanced and just at the time of fruit set, most eggs were laid on the leaves of the fruiting points. The number of eggs deposited on the fruit increased through the season until just prior to the normal harvest time. After reaching maturity, fruit attractiveness progressively decreased. More eggs were laid on twigs and wood in the beginning of the season than later on all cultivars checked. Hatting (1943) also noted differences in the number of eggs laid on the lower and upper leaf surfaces. On the cultivars 'Bon Chretien', 'Beurre Bosc' and 'Winter Nelis' more eggs were laid on the lower leaf surfaces. On the cultivar 'Glout Morceau', slightly more eggs were laid on the upper than on the lower leaf surfaces over the whole season. On the major pear cultivars currently cultivated on the Pacific Northwest the distribution of eggs has only been studied on the 'Bartlett' cultivar (Westigard et al. 1976). These authors indicated that the favored site for oviposition on this cultivar is the underside of fruit cluster leaves (74 % of the eggs found), and a low proportion of eggs (3-20 % depending on the year) was laid directly on the fruit. Since moths prefer the upper leaf surface on most apple cultivars, he hypothesized that this

preference may be due to the avoidance by females of the normally pubescent underside of the apple leaf. No information on egg distribution for winter cultivars, such as 'Anjou', is available. Similarly, Geier (1963) and Wearing et al. (1973) found that most eggs were laid within 10 cm of a fruit. Blomefield et al. (1997) found the majority of eggs within 12 cm of a fruit. MacLellan (1962) rarely found eggs more than 15 cm from fruit. These observations suggest that the search for a suitable oviposition site is directed as opposed to random and that certain cues guide a female to a fruit cluster.

Most studies did not show a significant effect of aspect on egg distribution, but MacLellan (1962) observed that early in the season eggs were mainly laid in the southeast portion on apple trees, but not later on the season.

Several authors have reported that codling moth damage is higher in the upper part of apple trees where mating takes place (Summerland & Steiner 1943, Woodside 1944, Geier 1963, Wood 1965). Blomefield et al. (1997) observed that differences in oviposition at different height of the tree depended on the time of year. Moths of the first generation appeared to prefer the bottom half and second and third generation moths the top half of the tree. No data is available about possible stratification of codling moth oviposition on pear cultivars.

2.8 Egg mortality

Most of the work done on egg mortality has focused on the effect of environmental variables and natural enemies as potential factors regulating codling moth populations. However, no information is available on host-specific egg mortality on different plant surfaces.

Hagley (1972a) determined that egg hatch was directly related to environmental humidity. Lekic (1950) stated that egg mortality occurred below 40 % relative humidity. Wyniger (1956) observed no egg hatch at 32 °C and less than 50 % relative humidity.

MacLellan (1962) determined that egg predation in an apple orchard accounted only for 14.4 % mortality and egg parasitism by *Trichogramma* sp. for 2.4 %.

In a field study by Ferro et al. (1975) on apples, egg mortality was 25 % under caged conditions, but increased to 50 % without cages. Westigard et al. (1976) reported 26% egg mortality in a 'Bartlett' pear orchard, with parasitism by *T. minutum* accounting for 10 % of the total.

2.9 Larval behavior

The searching behavior of neonate larvae (random or directed), as well as their physiological state, greatly influence their searching efficiency for fruit and, consequently, their survival rate (Jackson 1976, Jackson 1982). Geier (1963) distinguished between three activities of newly hatched codling moth larvae: (1) displacement to the fruit; (2) exploration of the fruit surface; and (3) penetration into the fruit. Another activity (4) feeding inside the fruit, can be added.

Several authors have studied the movement of neonate codling moth larvae towards the food source. McIndoo (1929) and Hall (1934) reported that neonate larvae move randomly after hatching. Other researchers thought codling moth orientation was more directed (Geier 1963). Sutherland (1972) found that, in the laboratory, newly hatched larvae moved rapidly in a klinotactic manner (directed movement towards the odor source through the comparison of concentrations of a chemical with side to side head movements) toward freshly cut apple skin. Sutherland (1972) and Sutherland & Hutchins (1973) found that the same behavior was elicited by two naturally occurring stereoisomers of alpha-farnesene, but that the sphere of attraction of these stimuli was only a few millimeters. However, the range of attraction of fruit odors in the field may differ from results obtained in the laboratory. Russ (1976) showed that neonate larvae preferred apple fruit extract as well as apple fruit as compared to pear extract or pear fruit. He hypothesized that this finding was related to a lower content of alpha-farnesene on pears. Bradley & Suckling (1995) and Landolt (1998) suggested that larval attraction to apple fruits could be related to orthokinetic responses (rate of locomotion change in response to changes of concentration of a chemical) to alpha-farnesene. When given a choice between previously infested and non-infested apples,

Jackson (1982) observed that fewer neonates chose the infested fruit when it had more than two entries, but there was no preference among non-infested apples and apples with one or two entries. Landolt (personal communication), on the other hand observed a preference for infested over non-infested apples in laboratory experiments using an olfactometer. McIndoo (1928) indicated that neonate larvae have a negative geotactic response and, consequently, they tend to move upwards. McIndoo (1928) and Jackson (1982) confirmed that first instar larvae are photopositive. Jackson (1982) observed that they were generally more active and moved faster in the light. Under field conditions, he observed that larvae typically moved rapidly for a short time after hatching before slowing down. Most larvae that had not located a fruit after 15 minutes crawled into protected places and became motionless, although later those same larvae successfully entered apples. McIndoo (1928) also indicated that first instar larvae use visual stimuli to locate objects, and smell was only useful within a few millimeters of the food source. Jackson (1982) studied the effect of temperature on first-instar behavior. Under laboratory conditions, larvae were unable to move at 5 °C or above 45 °C. The average speed over the rough surface of branches was 1.6 cm/min. The speeds of larvae were greater on smooth surfaces in the laboratory than on rough bark or pubescent leaf pedicels. Maximum average speed during 10-min trials was 4.5 cm/min at 30 °C. Larval pathways were less straight at higher temperatures. Steiner (1939) reported that first-instar larvae may travel long distances up to 3 m.

After reaching the fruit, larvae normally wander over the fruit surface for up to 2 h (Geier 1963). Jackson (1982) indicated that under laboratory conditions this wandering period was shorter (17 minutes).

Before breaking the epidermis of the fruit, a larva spins a few strands of silk on the surface of the fruit to ensure its footing and to anchor its body while chewing (Hoerner 1925, Hall 1929). Siegler (1940) and Chapman & Lienk (1971) indicated that young larvae bite off and discard the first pieces of epidermis. These authors hypothesized that cultivar differences in fruit penetration might result not only from differences in epidermal morphology of the fruit but also from other factors located in subepidermal layers of the fruit. Siegler (1940) also pointed to the importance of this habit as a factor that has

contributed to the difficulty in controlling codling moth. The time required for successful penetration varies considerably depending on temperature, time of day, fruit cultivar, and larval vigor (Hall 1929). Penetration times of 15 to 200 minutes have been reported (Hoerner 1925, Hall 1934). Larvae may have an easier access to the interior of the fruit through the calyx, or at cuts, stings, protuberances, lesions, or other irregularities of the fruit surface (Putman 1963). Provancher (1874) and Huard (1916) reported that codling moth larvae entered the fruit mostly by the calyx area. Mailloux & LeRoux (1960) observed that on apples larvae mostly preferred the side of the fruit as a point of entry. Fruits touching other fruits or leaves are more frequently attacked than a single fruit (Hall 1929, Audemard 1976). This is possibly a manifestation of thigmotactic behavior (Coutin 1959).

After penetrating, neonates initially feed near the surface of the fruit. After the first molt the larvae bore to the core where they consume the developing seeds and surrounding pulpy tissues (Jackson 1976).

Although normally a larva just attacks one fruit, Garlick (1940) observed that later instars sometimes leave an apple to enter another to complete development. This was noted mostly when apples were touching and when small fruits failed to develop. The movement from a fruit to another was mostly observed at night. Such behavior in first instar larvae would represent a drastic decrease on the probability of a successful penetration (Jackson 1982).

Several authors have observed the ability of young larvae to feed on food sources other than the fruit. Marshall (1940) noted that neonate larvae with no fruit available were able to feed on fruit buds and spurs, sap shoots, leaf bases, new breaks in twigs and small branches (4-year old wood), enlargements caused by the feeding of woolly aphids, and leaves alone. Spur-burrowing in 'Anjou' and 'Bosc' by codling moth larvae in an orchard in Oregon was reported by Gentner (1940). In some cases, leaf, bud and spur feeding may help sustain larvae during extended searching periods, but the frequency of this behavior is believed to be rare and the capability of the codling moth to reproduce when feeding only on food sources other than fruit has not been demonstrated.

Fifth instar larval behavior such as searching and selecting of a cocooning site can also play an important role in codling moth population dynamics. Various authors have studied the behavior of fifth instar larvae and the distribution of cocoons on tree. McIndoo (1928) indicated that fifth instar larvae are photonegative, geopositive, and thigmopositive. Garlick (1948) and Wearing (1975b) observed that larvae predominantly moved towards the main leaders and trunk away from the smaller branches. Most cocoons were formed under bark, in natural holes and depressions, and around pruning scars in the top of the tree. Another important percentage of cocoons was established on the main trunk. Fewer cocoons were found in the region of the main crotch, where the main trunk bifurcates. MacLellan (1960) found the trunk of apple trees the most preferred site for overwintering larvae to cocoon. Few cocoons are usually found on the ground. MacLellan (1960) indicated that the number of cocoons on the ground depended on the availability of cocooning sites on the tree and on the amount and type of debris on the ground. Garlick & Boyce (1940) and MacLellan (1960) reported that codling moth larvae seeking places to form the cocoon leave the fruit only during the hours of darkness. MacLellan (1960) considered this habit an adaptation to escape diurnal parasites and predators. In field experiments, larvae released at night were more successful in finding cocooning sites and they moved more regularly than larvae released during the day with little stopping for orientation or change of direction. He confirmed that fifth-instar larvae showed a photonegative response. He also noted that a high proportion of first and a low proportion of second generation larvae left the fruit before it fell to the ground.

2.10 Larval and pupal mortality

Since most codling moth eggs are laid on leaves in the vicinity of the fruits and not directly on them, newly hatched larvae must search for the fruits (Jackson 1979). If searching larvae do not reach food within a certain period of time, they stop moving and die of starvation. During searching neonate larvae may be exposed to attack by predators and parasitoids and to adverse weather conditions (wind, rain). The maximum time neonate larvae can search without feeding depends on how rapidly they exhaust their energy

reserves. Whether the search for a fruit is successful depends on the distance a neonate has to travel (distance between egg and fruit), the complexity of the intervening plant surfaces, microenvironmental variables (temperature, relative humidity, rain, wind), and the searching efficiency of neonate larvae. Larvae may die after reaching the fruit if they do not have enough energy to penetrate its epidermis (Jackson & Harwood 1980).

The studies that have been done on larval and pupal mortality focused mostly on the effect of environmental variables and natural enemies as potential factors regulating codling moth populations. In a very few instances host-specific mortality of immature stages has been associated with differences in host plant resistance.

Research has shown that the first (MacLellan 1971, Ferro et al. 1975, Jackson 1982) and the fifth larval instar (Garlick 1948, Wearing 1975a) have higher rates of mortality, because they are exposed to unfavorable environmental conditions and natural enemies. Cutright (1931) observed a positive correlation between temperature and the ability of neonate larvae to penetrate the fruit. At 15 °C no larvae entered the fruit while at 30 °C the fruit was covered with entrances. The same author also hypothesized that if the larva does not feed within two hours of hatching there is an average loss of weight of 12 % that stimulates action, which, in turn, increases the percentage of larvae established in the fruit. Jackson & Harwood (1980) observed in laboratory experiments that mean survival times of starved first instar declined as temperature increased and relative humidity decreased. At a constant relative humidity, survival time decreases as the temperature increases. At a given temperature, the average survival time increases as the relative humidity increases. Maximum survival time was over 200 h for neonate larvae in water-saturated air at 4 °C. Neonates survived less than 1 h in water-saturated air at 45 °C. The proportion of first instar larvae penetrating apples was reduced when larvae were starved for various lengths of time, especially at high temperatures (30 °C). Hagley (1972b) reported that mortality of codling moth larvae in apples under field conditions was highly correlated with total rainfall. The highest larval mortality (18.2 %) occurred when 1st instars were just beneath the epidermis of the fruit. Larval mortality was lowest (3.4 %) when 3rd instars were feeding in the center of the fruit. Rainfall during late spring and early summer significantly reduced the number of successful fruit entries. No information is available about differences in fruit

permeability to water that could partially explain differences in larval mortality on different host cultivars.

Ferro et al. (1975) determined that first instar mortality was 65 % before penetration. Total mortality of all larval stages inside the fruit was 35 % in apples. He also estimated that 5th instar mortality was around 30 % in an unsprayed orchard, although this value varied greatly depending on the impact of predators and parasitoids. MacLellan (1962) determined that mortality of overwintering larvae was 59 % in an unsprayed apple orchard. Among the more important mortality factors were bird predators, primarily woodpeckers, insect parasitoids (principally *Ascogaster quadridentata*), and low winter temperatures. Moisture conditions during overwintering have less effect on the survival of larvae than does the temperature. Prolonged frost may have a desiccating effect (Shel'Deshova 1967). Hagley (1969) indicated bird predation as the major mortality factor in overwintering larvae in southern Ontario. Furthermore, he observed differential overwintering larval mortality on different host cultivars. A smaller proportion of larvae appeared to survive the winter on the trunks of 'Spy' and 'Delicious' trees. He associated low survival with the smooth bark on the trunks of these cultivars that probably afforded the larvae less protection from parasitoids and predators. Wearing (1975a) also reported bird predation by the silvereye, *Zosterops lateralis*, as an important mortality factor in overwintering larvae in New Zealand. Bird predation was influenced by the distribution of cocoons and increased with the height in the tree and with placement in more accessible sites for parasitoids and predators (Wearing 1975b). Garlick (1948) studied the mortality of overwintering larvae, but he did not find any differences in two apple cultivars, 'Ontario' and 'McIntosh'.

Westigard et al. (1976) studied larval mortality on apple and several commercial pear cultivars. First instar larval penetration was higher on 'Golden Delicious' apple (> 70 %) than on any pear cultivar. Among the pear cultivars tested, mortality on 'Anjou', 'Bosc', and 'Comice', was higher than on 'Bartlett', especially during the middle of the season. The authors suggested that the low success of first instar to penetrate to the fruit interior on pears might be related to the presence of stone cells. While stone cells are absent from apple they are found to varying degrees in different pear cultivars. In southern Oregon, where the study was done, lignification of cells occurs normally in early June. Stone cells are present

until the fruit begins to ripen. This would explain why all pear cultivars are susceptible in early season, followed by a high degree of resistance to larval penetration in mid season, and finally by an increase in susceptibility to codling moth attack near harvest. Armstrong (1945) got similar conclusions from field observations on apple and pear orchards in Ontario.

Goonewardene et al. (1984) indicated that another factor responsible for resistance to codling moth injury could be the absence of seeds in the fruit. Seedless apples had lower infestation levels than fruit with a normal seed count. Larval survival in seedless fruit was significantly lower.

Ferro et al. (1975) found pupal mortality to be very low. Shelford (1927) indicated that considerable mortality of pupae could occur if relative humidity was less than 40%, although under optimum temperature conditions more than half of the pupae developed even when the relative humidity was 20-25 %.

2.11 Development of immature stages

The rate of development of the codling moth depends on temperature and, to a lesser extent, other abiotic and biotic factors (Glenn 1922, Shelford 1927, Gillot 1980).

The relationship between temperature and development of the codling moth has been the subject of several studies (Glenn 1922, Geier 1963, Shelford 1927, Hagley 1972a, Williams & McDonald 1982, Rock & Shaffer 1983, Setyobudi 1989). Although some of this research was conducted under natural, fluctuating temperatures, most of it was conducted under controlled laboratory conditions, using either artificial diet or apples as food source. Many of these studies included temperatures in the middle of the developmental rate curve, which can be reasonably approximated by a straight line. These authors showed that the accumulated thermal requirements for the development of a codling moth generation is about 600 degree-days and that the developmental thresholds and thermal requirements of different development stages are different. Although the lower developmental thresholds for eggs, larvae and pupae vary some a common lower threshold of 10 °C has been used for predicting codling moth phenology using a linear heat unit

system (Riedl & Croft 1978). According to Setyobudi (1989), the fourth larval instar has the highest lower threshold (14.2 °C) and the second instar has the lowest lower threshold (9.7 °C). The fact that male larvae have significantly lower threshold (10 °C) than females (11.3 °C) may explain why male codling moths appear first in the spring (about 2-5 days earlier than females) (Setyobudi 1989).

Later studies (Pitcairn et al. 1991, Serra 1994, Martí 1995, Martí et al. 1996) have shown that nonlinear models, like the one initially proposed by Logan et al. (1976), present a more accurate description of codling moth development. Non-linear models use specific thresholds for different immature stages and they predict the development more accurately when temperatures exceed the upper thermal threshold. Temperatures higher than the upper threshold not only stop development but also may slow it down. This would explain why linear methods predict a faster rate of development than the real one at temperatures near or above the upper threshold (Martí 1995, Martí et al. 1996). Pitcairn et al. (1991) reported that all stages failed to complete development at constant temperatures below 12.2 °C, and 14% of the eggs but no larvae or pupae survived at 34.4 °C.

The photoperiod has also been shown to influence postembryonic development. Russ (1976) noticed that codling moth larvae, after termination of diapause, developed faster to the pupal stage at longer photoperiods than in complete darkness. Setyobudi (1989) reported that a photoperiod of 14.75:9.25 (L:D) gives the fastest growth of codling moth larvae.

Larval development also seems to be affected by the food source, mostly by the stage of fruit development. Hall (1934), Gentner (1940), Marshall (1940), Heriot & Waddell (1942) and Cisneros & Barnes (1974) observed that immature fruits favor a faster larval development. Heriot & Waddell (1942) indicated that immature tissues have a greater nutritive value than mature tissues (both seeds and pulp). Mature tissues tend to increase larval longevity but decrease rate of development. Hathaway et al. (1971) also noted that developmental rates were shorter in individuals reared on immature apples than on several artificial diets. Developmental heat requirement may also vary among different codling moth host races. Phillips & Barnes (1975) determined that a plum race required less heat than an apple or walnut race to complete development.

2.12 Adult fecundity and egg viability

The reproductive capacity of a codling moth female depends on different factors, including the number of oocytes formed (potential fecundity), environmental factors, the ability to mate, food supply, longevity, and the availability of favorable oviposition sites (Geier 1963, Ferro & Akre 1975).

Newly emerged codling moth females contain 150-200 discernible oocytes in all stages of maturation (Wiesmann 1935). Several authors have indicated that potential fecundity varies with body weight (Geier 1963, Deseö 1971). Deseö (1971) reported that the fecundity of the codling moth female depends primarily on her weight and is influenced by the quality and quantity of larval nutrition. Wearing & Ferguson (1971), Deseö (1973) and Deseö & Saringer (1975a, 1975b) also noticed differences in fecundity among codling moth generations. They indicated that the overwintering codling moth generation having developed under field conditions has a very low average fecundity (17-23 eggs/female), in contrast to the first generation (76-83 eggs/female). Deseö (1973) associated this lower fecundity after diapause to several factors, including a lower proportion of males mating, a lower proportion of females laying eggs, and a reduction in the number of eggs laid per fertilized female after diapause (related to a lower body weight, among other factors). Geier (1963) related the low fecundity in females from the overwintering generation to the depletion of body reserves during winter dormancy. Lower fecundity was also observed in adults under pre-diapausing conditions (Deseö & Saringer 1975a, 1975b). They associated this alteration in the reproductive capacity of adults to the short photoperiod during egg and first instar larval development, and suggested it was independent of the processes related to the preparation for diapause.

Published estimates of the number of eggs laid per female vary widely and have been reviewed by Geier (1963) and Putman (1963). Hall (1929) obtained an average of 64 and a maximum of 234 eggs from spring-brood moths and an average of 83 and a maximum of 208 from first brood moths at Vineland Station. Geier (1963) reported a much lower mean of 44 and a maximum of 198 eggs per female in the Capital Territory. The greatest reported number of eggs laid by one female was 345 (Isley 1938). Much higher estimates of mean

fecundity have been obtained for some laboratory reared populations. For example, Gehring & Madsen (1963) reported mean fecundities over an 8-day period of 179.1 and 162.5 eggs/female for two different strains, respectively. Hagley (1972) obtained on average 236 eggs/female over a 15-day period. Putman (1963) indicated that field moths confined in the laboratory in a density of 1:1 (male:female) oviposit irregularly or not at all.

Deseő (1970) found no correlation between temperatures during larval and pupal development and the number of follicles and the number of eggs laid by adult females. The age of the female has also been reported as a factor influencing reproductive capacity. Vickers (1997) indicated that mean fecundity of females mated on the fourth day after emergence was significantly lower than in females mated either on the day of emergence or two days later. He, as Geier (1963), considered there is a trade-off between longevity and mating/fecundity.

The positive effect of adult nutrition on reproduction has also been reported. Wiesmann (1935) obtained twice as many eggs from sugar-fed females as from unfed individuals. Hamstead & Gould (1950) noted that fertilized females caged without water or food laid fewer eggs than did free-living moths, and suggested that starvation caused females to re-absorb part of their maturing oocytes. Several authors have studied the effect of nutrition of immature stages on adult fecundity. Speyer (1932) and Heriot & Waddell (1942) were successful in rearing moths from larvae fed on leaves alone. However, the moths were unusually small, were short-lived, and produced no eggs. Hathaway et al. (1971) reported that moths reared on apples were heavier, and consequently had higher reproductive potential, than those reared on artificial diets. Differences in reproductive capacity have also been noted when moths were caged in different apple cultivars. Wearing & Ferguson (1971) reported that moths caged on 'Red Delicious' apple trees laid more eggs than those on 'Jonathan', 'Sturmer Pippin', and 'King of Tomkin's County'. Whether differential fecundity potential exists among different pear cultivars has not been tested. Codling moth races adapted to different host plants have been shown to have different reproductive capacity. Cisneros & Barnes (1974) found that a walnut population had a higher reproductive potential than an apple population.

Other factors, such as the number of matings and the age of the female at the time of the first mating, reportedly affect production and egg viability. Deseő (1971) indicated that

the proportion of non-viable eggs increased with consecutive matings of the male (especially after their third and fourth copulation). She suggested that the low amount of reserves in the females was not the cause, and that the condition of the males probably influenced eggs viability. The ratio of eupyren and apyren spermatozoa could affect egg viability. The viability of the sperm could be affected by the inadequate functioning of the accessory sex glands in the males after consecutive matings.

2.13 Review summary and dissertation structure

The codling moth is an oligophagous species that attacks a variety of tree crops. It evolved from its original host, apple, to be able to attack and complete its biological cycle in other hosts, including other pome fruits, such as pear, but also some stone (apricot, plum), and nut fruits (walnut). There are large differences to codling moth attack among host species. Apple is in general more susceptible. There is a higher variability in susceptibility among pear cultivars. However, the literature offers little information about the factors associated with the observed differences in susceptibility. Differences in susceptibility among apple and pear cultivars may be associated with physical, anatomical, and chemical differences in hosts that can affect several aspects of the biology and behavior of the codling moth. Many aspects of codling moth biology, ecology, behavior, and control have been intensively studied on apple, its original and in general more susceptible host. However, the scope of codling moth research on pears has been limited and considerable gaps in understanding of codling moth biology and behavior remain. The question of how the adult stage of the codling moth finds a host tree, whether movement is random or directed in response to cues from host plants is not known. There is no information on the role of stimuli from host plants on codling moth mating. Egg distribution has been studied in several hosts, especially apple and some pear (e.g., 'Bartlett') cultivars. However, similar information is lacking for other commercial pear cultivars such as 'Anjou'. Factors affecting oviposition need to be studied in more depth to provide information about egg distribution on different host cultivars. Egg distribution on a host is indirectly related to susceptibility since neonate mortality can be expected to be higher in those cultivars where

eggs are laid farther away from the fruit. There is some information about how neonate larvae orient to a fruit and how mortality first-instar larvae while attempting fruit penetration may contribute to differences in susceptibility among hosts cultivars. However, cultivar-specific resistant factors which inhibit larval penetration have not been identified. The effect of larval food quality on the developmental rate and the reproductive potential of codling moth on apple and pear has not been studied. The heat requirements for completion of the development and the reproductive potential of codling moth on different hosts may not be the same. All the above mentioned aspects of the biology and behavior of the codling moth could contribute to the observed differences in susceptibility to codling moth attack among apple and pear cultivars.

The present work represents an effort to elucidate why codling moth is more successful on some hosts but not on others. Therefore, the research presented here focused on several key aspects of the relationship between the codling moth and its hosts that could explain the differences in susceptibility among hosts. The relationship between the different life stages of the codling moth and its hosts goes through a series of sequential steps including (1) host finding and selection by the adult stage after emergence, (2) female calling and adult mating, (3) oviposition by the female, (3) egg development and mortality, (4) fruit searching by neonate larvae, (5) fruit penetration attempt by first-instar larvae, (6) fruit consumption during larval development, (7) pupal development and adult emergence if conditions are favorable for continued growth and development, or diapause if they are not. The research followed the codling moth's life cycle which offered a logical sequence for planning and executing the experimental work. The aspects of the biology and behavior which were investigated here were organized in four main topics: (1) host selection by the adult stage, (2) mating and oviposition behavior, that can lead to a differential egg distribution in the field, (3) egg development and mortality and neonate larval behavior and mortality, and factors associated with resistance to larval penetration on apple and pear, and (4) effect of food source on larval and pupal development, adult fecundity, and egg viability. A separate thesis chapter is devoted to each topic. A concluding chapter summarizes the most important findings and points out areas for further research. A simple life table was generated in an attempt to summarize and quantify some of the factors which were

investigated here on the success of codling moth on specific hosts and on the infestation levels that can be expected in apple and pear cultivars.

Most of the experiments were conducted under field or semi-field conditions to obtain results that were representative of what happens in nature. It should be pointed out that in the field it is not possible to control all the variables that can affect the outcome of an experiment. Therefore, additional laboratory experiments were conducted to investigate specific aspects of the biology and behavior of the codling moth under controlled conditions.

CHAPTER 3
CODLING MOTH (LEPIDOPTERA: TORTRICIDAE)
HOST SELECTION

Santiago Martí, Helmut Riedl, and Jesús Avilla

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3.1 Introduction

The codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), is a worldwide pest of pome fruits, some stone fruits and walnuts (Shel'Deshova 1967, Riedl 1983). The severity of codling moth as a pest depends on the number of generations which are possible under local climatic conditions. In addition, hosts vary in terms of susceptibility to codling moth attack. Differences in infestation levels may be a manifestation of host preference by the moth stage and/or they may arise because of differential mortality of immature stages on different hosts. Apple, reportedly the original host of codling moth (Shel'Deshova 1967), is more susceptible than other hosts. Felt (1910) and Newcomer & Yothers (1924) observed differences to codling moth attack among apple cultivars and suggested that differences in infestation levels among early and late cultivars were related to the number of generations cultivars were exposed to in the field. Cutright & Morrison (1935) and Witehead (1944) suggested that other factors such as fragrance, skin hardness and sweetness of the fruit may also be related to codling moth susceptibility. In contrast to apple, there seems to be more variation in susceptibility to codling moth attack among pear cultivars. Westigard et al. (1976) reported that host-specific larval mortality could explain the variation in infestation levels observed among 'Golden Delicious' apple, 'Bartlett' pear, and winter pears, such as 'Anjou', 'Bosc', and 'Comice'.

Differences in infestation levels among host species and cultivars may also be a manifestation of host preference by the adult stage. Considering that codling moth is a specialist with a limited host range, it is likely that host finding and host selection is not a random process. Rather, moths may follow visual or olfactory/chemical cues to locate potential hosts. Codling moth adults may display a preference for a host over a non-host tree, for apple over pear or for a specific cultivar when given a choice. Larvae are probably not involved in the host finding and host selection process since their ability to disperse is limited.

How codling moth adults find a host tree, whether the search is random or directed, and whether long-range attraction is involved is not known. The literature offers little

information about these questions. When searching for a host plant, insects perceive and simultaneously integrate visual and chemical stimuli. Visual cues may include the spectral quality of the light stimuli, as well as the dimensions and shapes of the objects viewed (Schoonhoven 1968, Smith 1989). Host plants may release attractant and/or arrestment odors that can be used by the insect for recognition and orientation, while non-host plants may emit repellents and/or lack attractants and arrestment odors. The effect of host plant odors on insect behavior is receiving increasing attention. Responses of herbivorous insects to host plants may be strongly influenced by variations of host odor signal (Bäckman 1999). Release of volatiles may vary with the time of day and environmental factors such as temperature (Bäckman et al. 1998). Release of attractive volatiles by host-plants can be triggered by plant damage due to insect attack (Boevé et al. 1995, Turlings et al. 1997, Bernasconi et al. 1998), plant stress (Ebel et al. 1995), or mechanical injury (Landolt 1998).

There are few studies about the factors that aid codling moth adults to recognize, orient to and select a host plant. Laboratory studies have shown that exposure to apple odors increases locomotor activity of codling moth adults (Wearing et al. 1973, Buda 1978, Skirkevicius et al. 1980). Bäckman et al. (1998) reported increased electrophysiological activity in female antennae exposed to volatiles from apple fruits and branches with leaves and fruitlets. Skirkevicius & Tatjanskaite (1971), working with the codling moth attractant geraniol, determined that the response of adult moths to chemical stimuli was dependent on the time of day.

Codling moth adults can disperse over long distances. Marked moths have been trapped on host plants at distances up to 800 m from the point of release (Wildbolz 1958, White et al. 1973, Howell & Clift 1974, Suski et al. 1981) and some moths have been recovered over larger distances up to 8-11 km (Mani & Wildbolz 1977). However, none of these studies have given insight into possible cues affecting codling moth orientation to host plants.

The main objective of this study was to determine the role of visual and olfactory cues in guiding codling moth adults to host plants. Response of adult codling moths to host and non-host trees was investigated. Visual factors associated with the presence or absence of

host fruit, as well as size, shape, color, and density of fruit, and canopy silhouette, were investigated. The overall intention of this study was to assess whether host selection by the adult stage can explain observed differences in susceptibility to codling moth attack among apple and pear cultivars.

3.2 Materials and Methods

Codling moth populations. Moths for the experiments came from two sources. The majority of moths used in the release experiments were supplied by the Okanagan-Kootenay Sterile Insect Release (SIR) Program, Osoyoos, B.C. (Canada). This laboratory colony was originally collected in 1993 in an apple orchard in British Columbia (Canada) and has been maintained since on artificial diet (Brinton et al. 1969) at 27 ± 1.0 °C, 70-90% RH, and a photoperiod of 16:8 (L:D). Moths were sterilized with Cobalt 60 (25 krads) with a gamma cell 220. Calco Red 2144 (Passaic Color & Chemical Co., Paterson, NJ) was added to the diet that allowed easy field recognition of recaptured individuals. Adults were received weekly and kept in a cold room at 5 ± 1.0 °C and a photoperiod of 16:8 (L:D) h until their release. The second group of moths came from a laboratory colony collected in 1991 in an apple block at the Mid-Columbia Agricultural Research and Extension Center (MCAREC), Hood River, OR and was not sterile. This colony was maintained on thinning apples in an insectary at 24 ± 2.5 °C, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h (Riedl & Loher 1980).

Research sites and plant material. Field studies were conducted at two sites. The first research site was located at the MCAREC and consisted of a 0.2-ha mixed planting of 35-year old 'Anjou' and 'Bartlett' pear trees. Other apple and pear blocks surrounded this block. Additional six year-old 'Newtown' apple and three year-old maple, *Acer platanifolia* cv. 'Emerald Queen,' trees were planted between the pear trees. The apple trees were moved with a tree spade from an existing planting to the new site. The resultant rectangular block consisted of 15 trees of each tree type (Fig. 3.1). The distance between tree rows was 8 m and between trees in a row 4 m. Trees were pruned to make canopy

dimensions similar. Average height ($H \pm \text{SEM}$) and canopy width ($CW \pm \text{SEM}$) of the different tree species and cultivars were: ‘Anjou’: $H = 3.16 \pm 0.05$ m; $CW = 3.21 \pm 0.11$ m; ‘Bartlett’: $H = 3.13 \pm 0.04$ m; $CW = 2.85 \pm 0.05$ m; ‘Newtown’: $H = 3.15 \pm 0.04$ m; $CW = 2.74 \pm 0.06$ m; maple: $H = 3.11 \pm 0.07$ m; $CW = 2.12 \pm 0.06$ m. Similar experimental design was used by Prokopy et al. (1983) and Košťál & Finch (1994, 1996) to investigate the host selection of the cabbage root fly, *Delia radicum* (L.) (Diptera: Anthomyiidae).

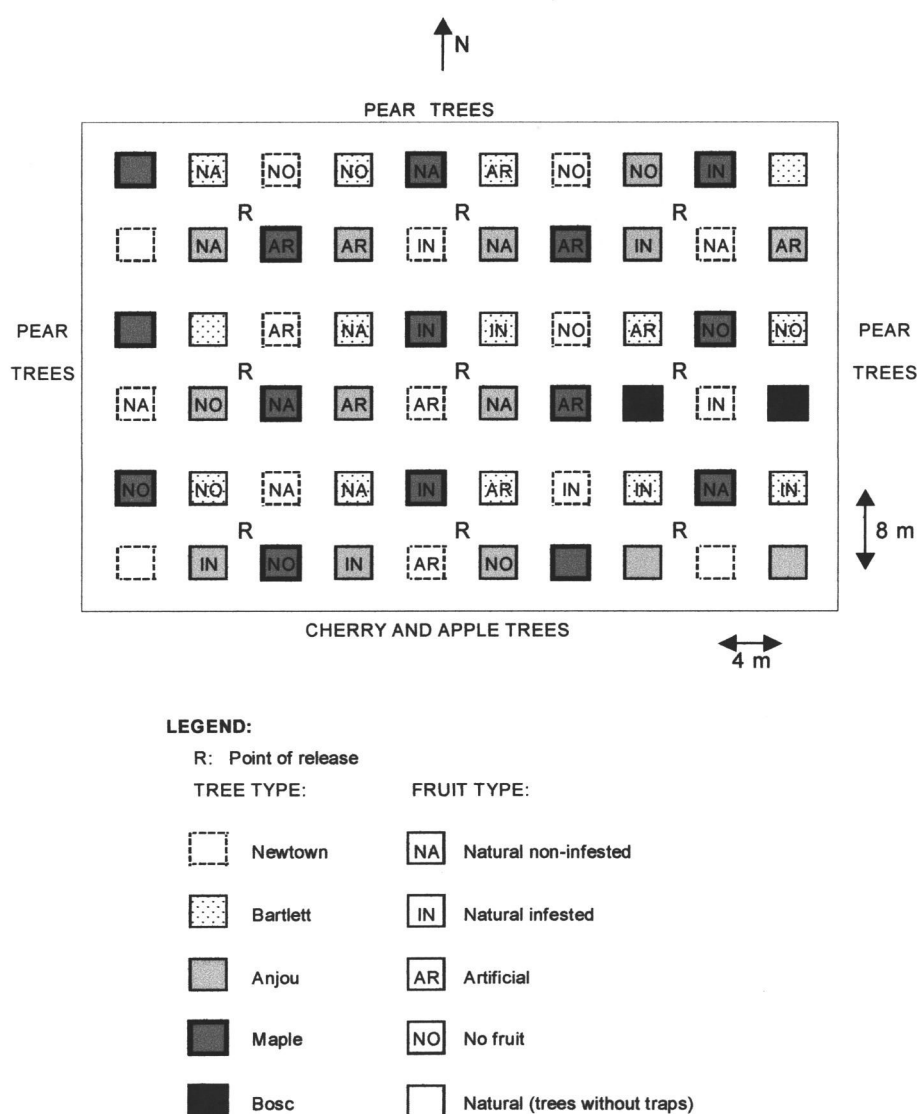


Fig. 3.1: Experimental layout at the MCAREC release site.

The second research site was a large open area (about 40 x 40 m) surrounded by commercial sweet cherry orchards located near The Dalles, OR. The Dalles site was chosen since it was several miles away from potential host trees and the release of fertile moths posed no threat to adjacent sweet cherry orchards. For the experiments, ‘Red Delicious’ apple, ‘Bartlett’ pear, maple, *A. platanifolia*, and artificial *Ficus* trees (H&P Sales Inc., Vista, CA) were placed singly in 75-dm³ containers and arranged in a circle. The apple, pear and maple trees were four, three, and two-year old, respectively. A total of nine trees of each natural tree type and 18 artificial trees were used in various arrangements in the different experiments. Examples for the layout of experiments of this site are shown in Fig. 3.2.

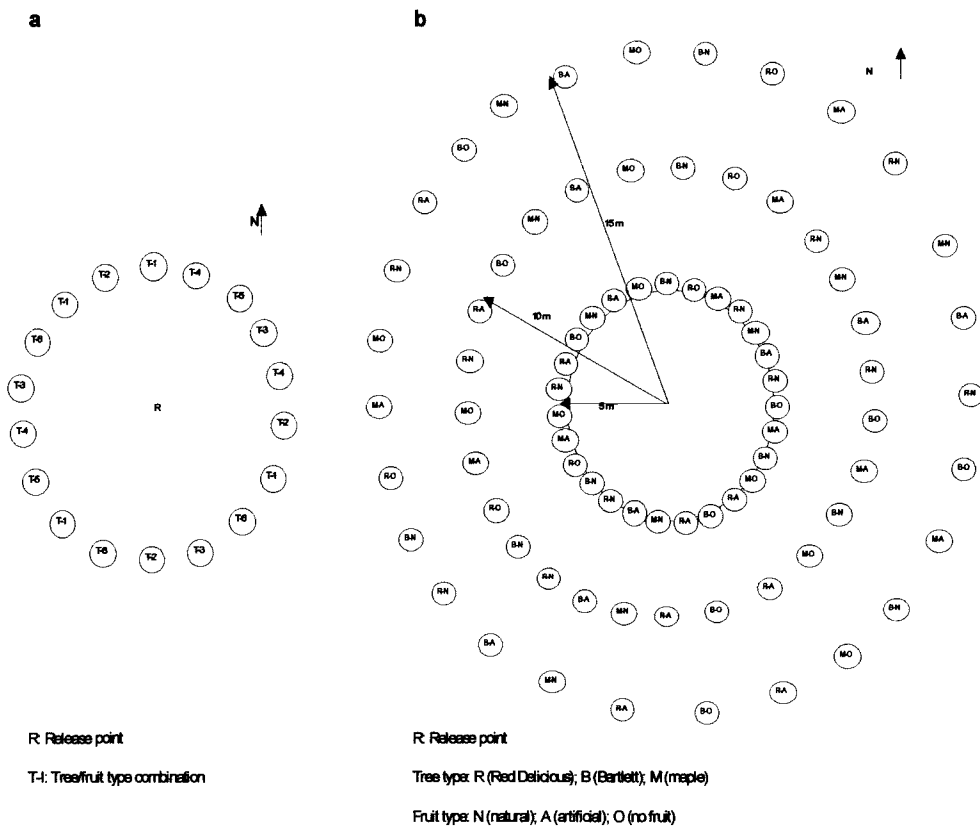


Fig. 3.2: Experimental layout at The Dalles release site: (a) general layout; (b) releases at different distances from potential hosts.

Average height ($H \pm \text{SEM}$) and canopy width ($CW \pm \text{SEM}$) of the experimental trees were: 'Red Delicious' apple: $H = 2.95 \pm 0.05$ m; $CW = 1.21 \pm 0.04$ m; 'Bartlett' pear: $H = 2.87 \pm 0.32$ m; $CW = 1.21 \pm 0.08$ m; maple: $H = 2.95 \pm 0.07$ m; $CW = 1.12 \pm 0.06$ m; maple: $H = 3.11 \pm 0.07$ m; $CW = 2.12 \pm 0.06$ m; and artificial trees: $H = 2.65 \pm 0.03$ m; $CW = 1.12 \pm 0.03$ m. Both natural and artificial fruit were used in experiments conducted at the two study sites. Artificial fruit measuring 18, 30 and 65 mm in diameter consisted of painted apple and pear-shaped objects made of Styrofoam® (Wang's International, Inc., Memphis, TN). Fruit was fastened on their petioles onto branches with tape.

Release and recapture procedures. All experiments were conducted during spring and summer of 1999 and consisted of multiple-choice tests. In the release experiments, the hypothesis were tested whether host selection by the adult stage of the codling moth is random or directed in response to specific cues (factors considered in different experiments) from host plants. Codling moth adults were released into different tree type/fruit type combinations. Sticky non-attractive interception pane traps were used to recapture released moths (Weissling & Knight 1994). Pane traps (0.3 x 0.3 m) consisted of clear, rigid, 3-mm thick acrylic plastic (Laird Plastic Co., Portland, OR) and were made sticky by painting both sides with a thin coating of STP 1075 Oil Treatment (STP Company, Fort Lauderdale, FL). Traps stayed transparent after application of STP and it was assumed moths were not able to perceive them visually (Weissling & Knight 1994). Holes were drilled in two corners of each pane, and wires were looped through each hole and attached to branches. At the MCAREC research site, traps (two per tree) were placed approximately at 2.5 m of height inside the canopy. One trap was placed in the east and the second one in the west quadrant of the tree. In experiments with potted trees, a single pane trap was placed in each tree at about 2 m. Moths were kept in a cold room at 5 ± 1 °C and were released between 5:00 and 7:00 p.m. Unless stated otherwise, each experiment involved four releases of 2400 individuals each. At the MCAREC site, since it was impossible to release all the moths at the same distance from all potential hosts, releases were made from several points (a total of nine) spaced evenly across the experimental block (Fig. 3.1). Each release point was 4.5 m away from the nearest trees. At The Dalles site, the situation was the ideal to conduct

multiple-choice preference tests with release moths. Releases were made from the center of the circular plot, so all moths were released at the same distance from all potential hosts. Release stations consisted of 1-m tall platforms on which a 1-liter cylindrical ice-cream carton (diameter 11 cm) with moths was placed. Two 10-cm long and 0.4 cm wide horizontal slits on opposite sides allowed moths to exit the holding container. The carton protected the moths from adverse weather conditions and bird predation. Moths and debris were removed daily from the sticky pane traps with forceps to avoid loss in trapping efficiency. After each release, traps were checked daily for three consecutive days. No recaptures were expected the fourth and subsequent days after release (Weissling & Knight 1994). In each experiment, traps were cleaned and re-coated before moth release.

Effect of tree and fruit type on host selection through the season. In this two-factorial experiment at the MCAREC site, the hypothesis was tested that host selection by the adult stage of the codling moth is a directed response to specific cues from foliage and fruit. The effect of presence or absence of fruit on the response of moths to tree type at different times during the season was investigated. Four levels of tree type ('Anjou', 'Bartlett', 'Newtown', and maple) were combined with four levels of fruit type (natural non-infested fruit, natural infested fruit, artificial fruit, and no fruit; Fig. 3.1). Three trees of each host and non-host were randomly chosen for each tree type/fruit type combination. The experiment was repeated four times during the season. The first test was conducted in mid-June (11-13, 14-16, 18-20, and 21-23), the second during the first half of July (June 28-30, and July 5-7, 9-11, and 12-14), the third in mid-August (Aug 10-12, 13-15, 17-19, and 20-22), and the fourth during the first half of September (4-6, 8-10, 11-13, and 14-16). Four releases were made during each time period. Corresponding to conditions in the field, the size of the artificial fruit used in the experiments at different time periods matched natural fruit growth. Fruit diameters were about 18, 30, 65 and 65 mm in the four sets of experiments from early to late season, respectively. Fruit densities were adjusted to 150, 100, 60, and 50 fruit/tree in successive experiments to reflect the normal decrease in fruit density over the season. Trees were thinned by hand to obtain the desired fruit densities. Blossoms were removed on trees that were chosen to have artificial or no fruit. Fruits were artificially infested with neonate larvae from the MCAREC laboratory colony. Due to

differences in larval penetration success, infestation levels varied with tree type and time of season. Average infestation levels on 'Anjou' trees were about 20, 40, 50 and 50 % in experiments one to four, respectively. Infestation levels on 'Bartlett', 'Newtown' and maple were about 40, 50, 70, and 80 % in each release period. Statistical analyses were carried out separately for each release experiment because of the different experimental conditions.

Effect of tree, fruit, and canopy type on host selection. In this three-factorial experiment at The Dalles site the combined effects of tree, foliage, and fruit on host selection by codling moth adults were investigated. The experiment combined two tree types ('Red Delicious' apple, 'Bartlett' pear), two types of foliage (natural, artificial) and two fruit types (natural, artificial). Each combination of tree, foliage, and fruit type was represented in the experiment by three trees. Potted trees were arranged at random in a 20-m circle (Fig. 3.2a). Moths were released from the center of the circle. Each release consisted of 1600 adults. Fruits (diameter 30 mm) were placed in trees at a density of 20/tree. The leaf to fruit surface ratio was approximately 10:1. Leaf surface was measured with a portable 3000-LI area meter (LI-COR, Inc. Lincoln, NE). Fruit surface was approximate to the surface of a sphere of the same diameter. The experiment was repeated four times (July 9-11, 12-14, 16-18, and 19-21).

Effect of tree and fruit type and distance on host selection response. Host selection behavior was studied with moths released at three different distances from potential hosts. Each release distance involved a separate experiment. Host selection responses over longer distances could not be studied because of space constraints. At each distance, codling moth adults were given a choice of three tree types ('Red Delicious' apple, 'Bartlett' pear, maple) with three fruit types (natural, artificial, no fruit). Each combination of tree/fruit type was represented in the experiment by three trees. Trees were arranged at random in a circle at The Dalles site (Fig. 3.2b). Since the number of trees was fixed, the distance between trees increased with the distance to the point of release (5 m → 1.2 m; 10 m → 2.3 m; 15 m → 3.5 m). Artificial and natural fruit (diameter 30 mm) was placed in trees at a density of 20/tree. The experiment was carried out with potted trees and repeated four times for each distance: June 23-25 and 27-29, and July 1-3 and 4-6, for 5 m; August

24-26 and 29-31 August, and September 1-3 and 4-6, for 10 m; August 10-12, 13-15, 17-19, and 21-23, for 15 m.

Effect of canopy shape on host selection. A release experiment was conducted at The Dalles site to examine if the shape of the canopy affected host selection by the codling moth. Artificial trees were manipulated to obtain canopies with columnar (linear), pyramidal (triangular), and spherical (round) silhouette. Six trees of each shape were arranged at random in a circle (diameter 20 m). Trees had no fruit. Moths were released in the center of the circle. Each release consisted of 1600 adults. The experiment was conducted with potted trees and repeated four times in the spring: May 28-30, and June 1-3, 11-13, and 14-16.

Effect of fruit size, shape and color on host selection. The role of fruit size was studied in an experiment with potted trees using a two-factor arrangement with three tree types ('Red Delicious' apple, 'Bartlett' pear, maple) and artificial pear fruit of three different sizes (18, 30, and 65 mm). The experiment was conducted at The Dalles site. Two trees were used for each combination of tree type and fruit size. Twenty fruit were placed in each tree. Trees were arranged at random in a 20 m circle. Each release consisted of 1600 adults. The experiment was repeated four times: June 18-20, 25-27, and 28-30, and July 5-7.

The hypothesis was tested that host selection by the adult stage of the codling moth is a directed response to visual factors (shape and color) of host fruit in another experiment with potted artificial trees using apple- or pear-shaped artificial fruit painted red, green, or yellow. The red paint was a tartar dark red enamel (F65R2; Sherwin-Williams Co., Cleveland, Ohio). Green and yellow paints were obtained by mixing a semi-gloss yellow latex enamel base paint (AXX: Hearst Corp., Wal Mart Stores, Bentonville, AR) with different colorants which included: B: lamp black; C: yellow oxide; KX: titanium white; D: thalo green; and F: red iron oxide. For the green paint, colorants were added to the base paint in the following proportions (volume): AXX: 2.25; D: 0.08; F: 0.03; KX: 0.16; and for the yellow paint: AXX: 0.47; B: 0.02; C: 1.17; KX: 0.31. Fruit size (diameter 65 mm) and fruit density (15 fruits/tree) was the same in all trees. Two trees were used for each

combination of fruit shape and color. Trees were randomly arranged in a 20-m circle. Moths were released in the center of the circle. The experiment was repeated four times on September 7-8, 9-11, 12-13, and 13-15.

Effect of fruit density on host selection. Adult recapture on trees with four different fruit densities (0, 5, 10, and 20 fruits/tree) was compared to test the effect of fruit density on host selection. Artificial trees with natural 'Red Delicious' apples (diameter 60 mm) were used in this experiment. Three trees were used for each fruit density. Trees were arranged at random in a 20 m circle. Moths were released from the center of the circle. The experiment was repeated four times on September 15-17, 18-20, 21-23, and 24-26.

Host selection behavior of different codling moth populations. The host selection response of codling moth adults from two different populations was compared using a factorial design with three tree types ('Red Delicious' apple, 'Bartlett' pear, maple) and three fruit types (natural fruit, artificial fruit, no fruit). Three trees were assigned to each tree/fruit type combination. Fruit size (diameter 30 mm) and fruit density (20/tree) were the same in trees with natural or artificial fruit. The two strains in the test were the sterile moths from Canada and the MCAREC laboratory population. Moths of both strains were released simultaneously. The experiment consisted of four separate releases. Each time 2400 moths of the Canadian population were released and 100 or 150 moths of the MCAREC population. MCAREC moths were marked with yellow fluorescent powder (DayGlo Color Co., Cleveland, OH) to distinguish them from the sterile Canadian and feral moths. Fluorescent powder was added to the interior of ice-cream cups (11 cm diameter). The powder was distributed through the interior walls of the cup. Moths were put inside the cup for 5 minutes. During this time cup was moved so moths were better impregnated with the powder.

Determination of fruit and leaf characteristics. Content of α -farnesene in fresh leaf and fruit tissue of host trees ('Red Delicious' apple, 'Anjou' and 'Bartlett' pears) and fresh maple leaf tissue was determined at six different times during the 1998 season. Fruit ranging in weight from 2 to 170 g (diameter 10 mm to 65 mm) and leaves (10 g per sample)

from both fruit and non-fruit clusters were randomly selected for hexane extraction. Leaf and fruit samples were individually weighed. Leaf surface area was determined with a portable LI-3000 leaf area meter (LI-3000, LI-COR, Inc. Lincoln, NE). Fruit surface was approximate to the surface of a sphere of the same diameter. Samples were dipped in 100 ml HPLC-grade hexane for 3 min, turning the fruit and leaves manually during dipping (Meir & Bramlage 1988). Aliquots of the hexane extracts were diluted appropriately and scanned for UV absorption with a spectrophotometer (Model DU-64, Beckman Instruments, Fullerton, CA) between 190 nm and 300 nm wavelengths. Absorbance was recorded at 2 nm intervals. The mean absorbance of three replicates per treatment was plotted against the corresponding wavelengths at 2 nm intervals to obtain the UV spectrum for each treatment. Alpha-farnesene was measured at O.D. (=absorbancy) = 232 (molar extinction coefficient: $E_{232} = 27,700$) and was expressed as nano-moles per cm^2 of fruit or leaf.

Natural and artificial leaves from host and non-host trees, painted artificial fruit, and natural host fruit (20 and 50 mm of diameter) were analyzed with a Minolta CR-300 colorimeter (Minolta Co., Ltd, Osaka, Japan) to determine their reflectance in the visible spectrum and color properties (hue, lightness, and saturation).

Measurement of environmental variables. Temperature and wind speed were recorded at both study sites with a CR10 weather station (Campbell Scientific Inc., Logan, UT). Daily precipitation data were obtained from NOAA (National Oceanic and Atmospheric Administration) weather stations at The Dalles and at the Mid-Columbia Agricultural Research and Extension Center in Hood River.

Statistical Analysis. Combined codling moth captures per tree during a three-day period after release was the variable used for statistical analysis. Statistical procedures, such as two-sample *t*-tests, analysis of variance (ANOVA), and linear contrasts for mean comparisons using Waller-Duncan Bayes LSD test, were carried out with the Number Cruncher Statistical System (Hintze 1997). Data were transformed to $\log(x+1)$ for the analysis. These analyses were supplemented by non-parametric Friedman tests. Untransformed means ($\pm$ SEM) are reported in the figures.

3.3 Results

General observations: trap recapture efficiency and weather effect on recapture.

Moth recapture efficiency of interception traps was very low. On average, 1.91% of the sterilized Canadian moths and 8.75% of the non-sterile MCAREC moths were recaptured.

The environmental conditions during the experiments affected codling moth recapture. No moths were caught on the sticky pane traps when hourly average temperatures during the period of flight activity (between 7:00 and 11:00 p.m.) were lower than 12 °C. These conditions were an important factor in codling moth recapture at the beginning (May-early June) and at the end (September) of the season. Rain inhibited codling moth flight during the experiments and it was a big factor decreasing codling moth recapture. Wind was not a critical factor for codling moth flight. Wind speeds up to 10 km/h did not inhibit codling moth flight. Wind speed ranged from 0 to 35 km/h during the experiments. Maximum hourly wind speed reached values of 10 km/h or higher about 40 % of the evenings during the time period experiments were conducted.

Moths were recaptured during the three-day period after release. However, recaptures dropped significantly to very low levels after the second day after release ($F = 37.51$; $df = 2,6$; $P < 0.001$; Fig. 3.3). Only when the average temperature during codling moth flight activity period was below about 16 °C during the first or the two first days after release, the number of recaptures was similar during the three day-period catches were recorded (Fig. 3.4). However, under these conditions, recapture rates were very low.

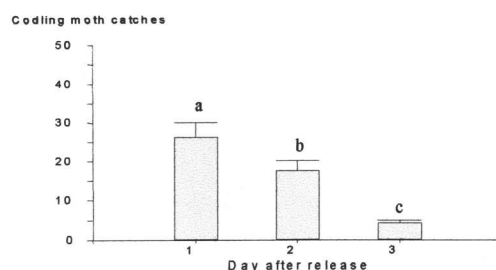


Fig. 3.3: Average number of codling moth recaptures across all treatments in successive days after release. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

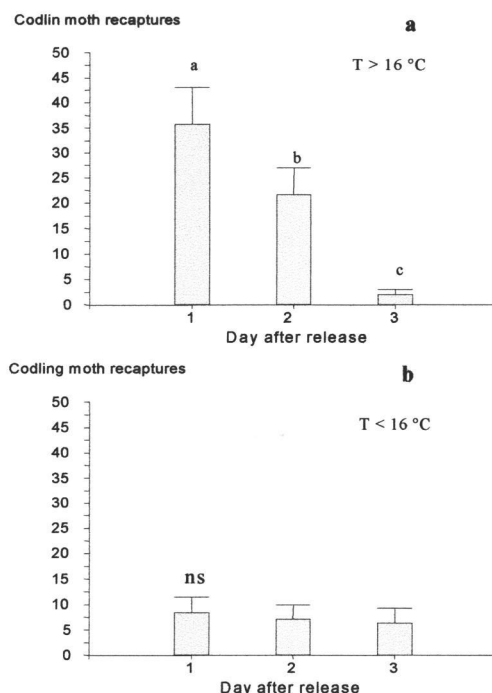


Fig. 3.4: Average number of codling moth recaptures across all treatments in release experiments when average hourly temperature from 7:00 to 11:00 p.m. was (a) $>16^{\circ}\text{C}$, and (b) $<16^{\circ}\text{C}$ the first two days after release. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Significantly more males (sterilized: 3.06%; non-sterilized: 10.67%) than females (sterilized: 0.76; non-sterilized: 6.83%) were recaptured in the experiments (ANOVA: testing significance of variable 'sex'; $P < 0.05$). Recapture efficiency at the two research sites was similar. At the MCAREC site 3.11% males and 0.76% females and at The Dalles site 3.02% males and 0.75% females were recaptured. However, male and female moths responded in a similar manner to tree/fruit type combinations (ANOVA testing significance of interactions among the variable 'sex' and all other factors; $P > 0.05$).

Response of male and females to different treatments was consistent on the three consecutive days after release in all replications of each experiment (ANOVA: testing significance of interactions among 'day after release' variable and all other factors; $P > 0.05$). Therefore, combined captures of both sexes per tree during a three-day period after release was the variable used for statistical analyses.

Effect of tree and fruit type on host selection responses through the season. From the results of the experiment at the MCAREC site, stimuli from the canopy (tree type) seemed to play an important role during the whole season (June: $F = 23.54$; $df = 3, 9$; $P < 0.001$; July: $F = 23.59$; $df = 3, 9$; $P < 0.001$; August: $F = 38.86$; $df = 3, 9$; $P < 0.001$; September: $F = 8.17$; $df = 3, 9$; $P = 0.006$; from ANOVA testing the significance of the variable tree type). Trees of host cultivars were significantly preferred over non-host trees (maple). Among host trees, pear trees were always more attractive than ‘Newtown’ apple trees. Among pear cultivars, moths showed no significant preference for ‘Bartlett’ over ‘Anjou’ trees, except at the end of the season when ‘Bartlett’ was preferred (Fig. 3.5).

The role of fruit stimuli on the host selection response of moths varied through the season but was consistent among cultivars. Early in the season, when the fruit was still small (diameter ca. 18 mm), moths were recaptured in equal numbers in host trees with natural non-infested, natural infested, artificial, and no fruit ($F = 0.53$; $df = 3, 9$; $P = 0.673$; Fig. 3.5). However, in subsequent release experiments visual and/or olfactory cues associated with fruit significantly enhanced the response of codling moth adults to host trees (ANOVA: testing for significance of fruit type in July: $F = 40.96$; $df = 3, 9$; $P < 0.001$; August: $F = 16.83$; $df = 3, 9$; $P < 0.001$; September: $F = 8.82$; $df = 3, 9$; $P = 0.005$). Trees with natural and artificial fruit were preferred over trees with no fruit (Fig. 3.5). Trees with natural fruit (non-infested and infested) were favored over trees with artificial fruit, especially late in the season (August and September) when fruit approached maturity (Fig. 3.5). Moths showed a similar preference for trees with natural non-infested and natural infested fruit during the whole season (Fig. 3.5).

The relative importance of cues associated with foliage and fruit was consistent among tree types. The ANOVA showed no significant interaction between tree type and fruit type (June: $F = 0.85$; $df = 9, 27$; $P = 0.575$; July: $F = 2.25$; $df = 9, 27$; $P = 0.050$; August: $F = 1.87$; $df = 9, 27$; $P = 0.100$; September: $F = 0.78$; $df = 9, 27$; $P = 0.640$). However, the interaction term tree type x fruit type in July and August approached significance at the $P = 0.05$ level.

At the end of the season, trees of the ‘Bartlett’ cultivar with natural non-infested and infested ripe fruit were equally favored as hosts and preferred over trees with artificial fruit (Fig. 3.5).

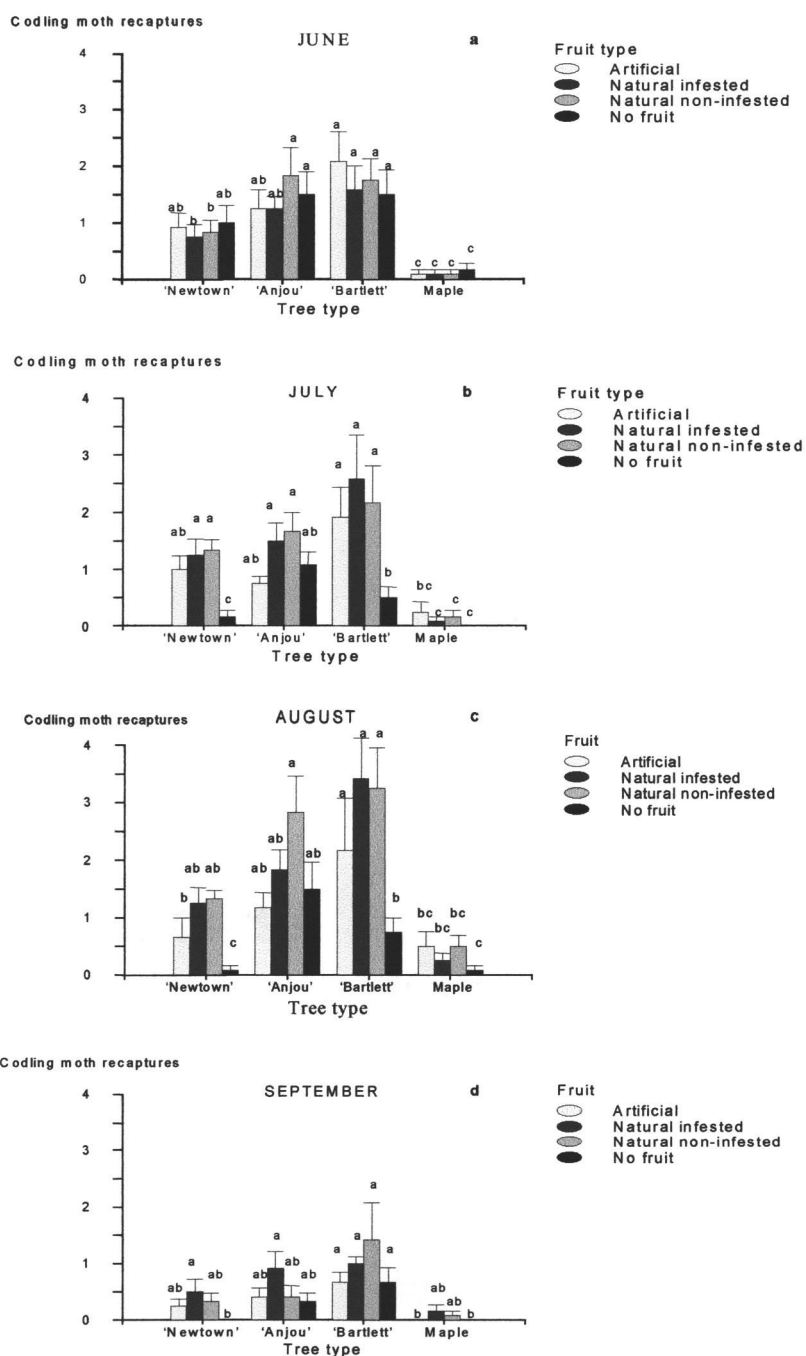


Fig. 3.5: Codling moth adult recaptures on different tree types with different fruit types through the growing season: (a) June; (b) July; (c) August; and (d) September. Each value is the combined male and female mean over the three-day period after release for each tree/fruit type combination. For a period during the season, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Role of α -farnesene and other plant volatiles in host selection. Alpha-farnesene is released by pome fruits (apple, pear) and reportedly affects oviposition behavior of codling moth females (Sutherland et al. 1974) and searching behavior of neonate larvae (Landolt et al. 1998). The content of α -farnesene in fresh leaf and fruit tissue of host trees ('Red Delicious' apple, 'Anjou' and 'Bartlett' pears) and fresh maple leaf tissue was determined at six different times during the 1998 season is presented in Table 3.1.

Table 3.1: Alpha-farnesene content on leaves and fruits of host cultivars and leaves of maple.

Plant material	Cultivar	Alpha-farnesene content (nmoles/cm ²): Mean (SEM)					
		May 11-12	June 05-06	July 07-08	Aug. 12-13	Sept. 03-04	Sept. 21-22
Fruit	'Red Delicious'	15.41 (0.76)	1.60 (0.32)	1.32 (0.13)	1.14 (0.09)	1.84 (0.52)	0.51 (0.12)
	'Anjou'	7.74 (0.68)	2.70 (0.19)	4.85 (1.57)	2.00 (0.22)	0.94 (0.11)	1.47 (0.75)
	'Bartlett'	9.45 (0.80)	2.21 (0.21)	3.62 (1.41)	1.60 (0.09)	0.94 (0.47)	2.12 (0.70)
Fruit-cluster leaves	'Red Delicious'	0.50 (0.01)	0.53 (0.01)	0.43 (0.01)	0.42 (0.03)	0.36 (0.06)	0.20 (0.01)
	'Anjou'	1.23 (0.03)	0.96 (0.03)	0.72 (0.02)	0.74 (0.09)	0.74 (0.06)	0.55 (0.02)
	'Bartlett'	0.58 (0.02)	0.80 (0.02)	0.56 (0.02)	0.59 (0.05)	0.45 (0.02)	0.31 (0.03)
No fruit-cluster leaves	'Red Delicious'	0.51 (0.02)	0.37 (0.02)	0.37 (0.04)	0.24 (0.02)	0.24 (0.04)	0.17 (0.02)
	'Anjou'	1.36 (0.07)	0.88 (0.02)	0.80 (0.05)	0.63 (0.05)	0.50 (0.02)	0.63 (0.09)
	'Bartlett'	0.63 (0.02)	0.76 (0.03)	0.51 (0.02)	0.49 (0.05)	0.27 (0.02)	0.44 (0.03)
	Maple	0.03 (0.00)	0.02 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)

The content of α -farnesene was higher in fruits (0.9-15.4 nmoles/cm²) than in foliage (0.2-1.4 nmoles/cm²) of host cultivars during the whole season. Among host cultivars, α -farnesene content was higher in 'Anjou' and 'Bartlett' pears than in 'Red Delicious' apple leaves and fruits. However, content was higher in 'Red Delicious' fruits (15.4 nmoles/cm²) than in pear fruits (7-9 nmoles/cm²) at the beginning of the season. Among pear cultivars, α -farnesene content was higher in 'Anjou' than in 'Bartlett' leaves and fruit during the whole season. Alpha-farnesene content in fruit was higher at the beginning of the season and reached a peak at mid-season. Alpha-farnesene content in foliage tended to decrease through the season in all host cultivars tested. Content in non-host maple leaves was insignificant (almost zero) during the whole season.

Visual characteristics of foliage and fruit and their role in host selection. Codling moth adults may also be able to discriminate among host trees because of differences in visual characteristics of foliage and fruit. Color attributes and reflectance curves of artificial and natural plant surfaces are shown in Table 3.2 and Fig. 3.6, respectively. Artificial fruit and foliage have similar reflectance curves and color attributes than natural surfaces of the same color. Fruits have twice the reflectance of the foliage. For both, reflectance begins to increase at 500 nm and peaks at about 550 nm. Yellow 'Bartlett' fruit reflects light in the same part of the spectrum but at greater intensity: 60 % at 550 nm versus 35 % of green 'Anjou' and 'Bartlett' fruit, and about 20 % for the foliage at that wavelength. Reflectance of red fruit, such in mature 'Red Delicious', begins to increase at 580 nm. Big 'Newtown' fruit, as well as small apple and pear fruit, has both green and red colors properties. Foliage of different plant species and cultivars has very similar reflectance patterns. Reflectance peak of artificial foliage is at 520 nm versus 550 nm of natural foliage.

Table 3.2: Color attributes of foliage and fruit of four different host cultivars, foliage of non-host maple, and artificial foliage and fruits used in experiments. L: lightness (pale/dark scale); C: chroma or vividness (gray scale); h: hue or color.

Plant organ	Tree type	Surface	L	C	h
Leaf	Maple	Underside	43.86	21.64	163.33
		Upper side	42.30	22.26	153.05
	Bartlett	Underside	50.80	29.54	110.23
		Upper side	43.04	29.25	113.88
	Anjou	Underside	53.51	23.39	113.21
		Upper side	39.55	21.53	117.07
	Newtown	Underside	52.90	22.86	112.80
		Upper side	38.80	20.54	118.36
	Red Delicious	Underside	52.82	23.80	111.06
		Upper side	38.23	20.51	118.96
	Artificial	Underside	50.78	21.03	111.25
		Upper side	33.44	13.17	121.79
Small fruit	Bartlett		36.80	20.13	80.72
	Anjou		38.21	18.54	62.61
	Newtown		46.92	24.52	85.61
	Red Delicious		51.75	33.06	88.59
Big fruit	Newtown		64.29	41.29	79.86
	Red Delicious		31.87	27.08	22.22
	Bartlett	Yellow	72.36	48.99	81.26
		Green	59.50	37.72	101.70
	Anjou		57.05	35.02	101.03
	Artificial	Yellow	65.03	47.16	79.07
		Red	34.23	35.05	23.10
		Green	57.84	41.20	101.06
		Green (no paint)	70.55	55.60	104.23

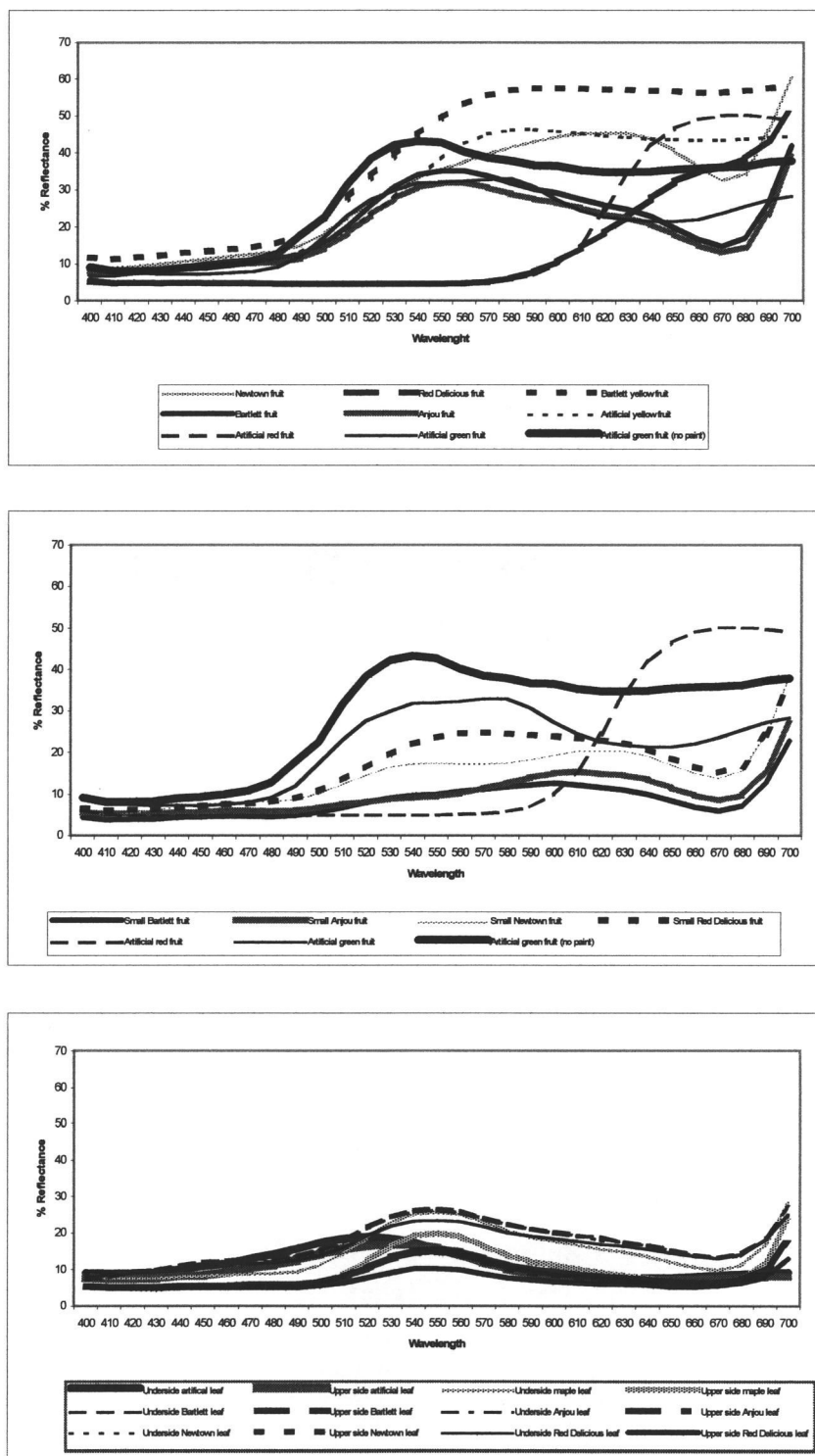


Fig. 3.6: Reflectance curves for foliage and fruit of four different host cultivars, foliage of non-host maple, and artificial leaves and fruits used in experiments.

Role of olfactory stimuli from canopy and fruit on host selection. Codling moth adults were equally attracted to ‘Red Delicious’ apple and ‘Bartlett’ pear trees ($F < 0.01$; $df = 1, 3$; $P = 0.953$). Trees with natural foliage were preferred over artificial trees ($F = 34.12$; $df = 1, 3$; $P = 0.010$). Moths were more attracted to trees with natural fruit than trees with artificial fruit ($F = 46.10$; $df = 1, 3$; $P = 0.006$). No interaction between tree and fruit factors was observed ($F = 1.22$; $df = 1, 3$; $P = 0.349$). Artificial trees with natural fruit were not significantly preferred over trees with natural foliage and artificial fruit (Fig. 3.7). There was some additive effect of olfactory stimuli from natural canopy and natural fruit (Fig. 3.7).

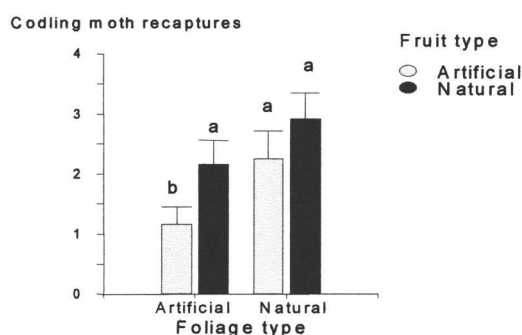


Fig. 3.7: Codling moth adult recaptures on artificial and natural trees having either natural or artificial fruit. Each value is the combined male and female mean over the three-day period after release for each foliage/fruit type combination. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Role of visual and olfactory stimuli from plants at different distances on host selection. Codling moths released at 5, 10, and 15 m from potential hosts responded in a similar way. Adult moths showed a significant difference on preference as host among tree types (ANOVA testing interaction distance x tree type: $F = 7.56$; $df = 2, 6$; $P = 0.023$) and among fruit types (ANOVA testing interaction distance x fruit type: $F = 82.53$; $df = 2, 6$; $P < 0.001$) from all distances. When released from 5 and 10 meters, moths behaved the same.

Adult moths significantly preferred apple and pear trees over maple trees; they were more attracted to trees with natural fruit than to trees with artificial fruit, and trees with no fruit were significantly less preferred (Fig. 3.8). When released from 15 meters a similar pattern was observed, but only 'Red Delicious' trees, not 'Bartlett', were significantly preferred over maple trees (Fig. 3.8). Moreover, trees with natural fruit were significantly preferred over trees with artificial fruit and trees with no fruit. The addition of artificial fruit did not significantly increase the number of catches compared with trees with no fruit (Fig. 3.8). These observations explained the significance of the interaction term among the factors tree and fruit type ($F = 13.22$; $df = 4, 12$; $P < 0.001$).

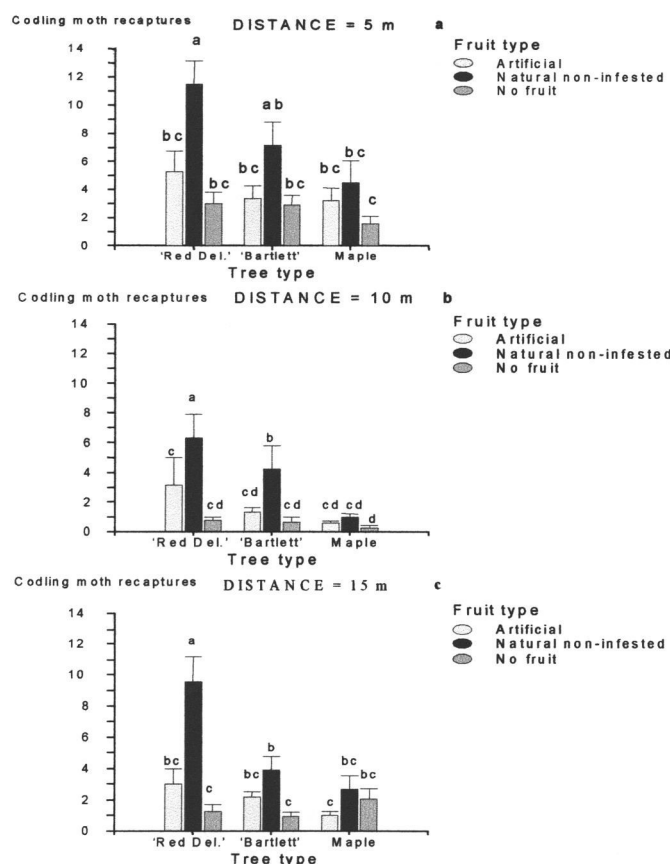


Fig. 3.8: Codling moth adult recaptures on different tree types with different fruit types when released at different distances from potential hosts: (a) 5 m; (b) 10 m; and (c) 15 m. Each value is the combined male and female mean over the three-day period after release for each tree/fruit type combination. For each distance, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Effect of visual cues from canopy and fruit on host selection. In Experiment 1, codling moth adults did not discriminate among artificial trees with different silhouette (columnar, triangular, round) ($F = 2.70$; $df = 2, 6$; $P = 0.145$; Fig. 3.9).

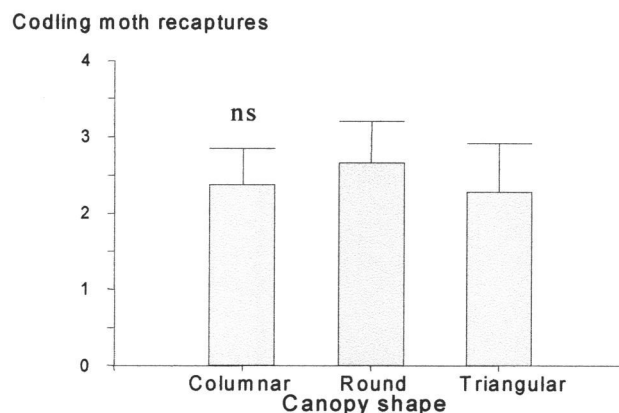


Fig. 3.9: Codling moth adult recaptures on artificial trees with different shape (silhouette). Each value is the combined male and female mean over the three-day period after release for each tree type. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

In Experiment 2, trees with bigger fruit (65 mm diameter) were significantly preferred as host over trees with medium (30 mm of diameter) and small (18 mm of diameter) fruit ($F = 5.74$; $df = 2, 6$; $P = 0.040$; Fig. 3.10). Apple and pear trees were favored as host over maple trees ($F = 10.91$; $df = 2, 6$; $P = 0.010$). There was no interaction between the factors tree type and fruit size ($F = 0.18$; $df = 4, 12$; $P = 0.944$).

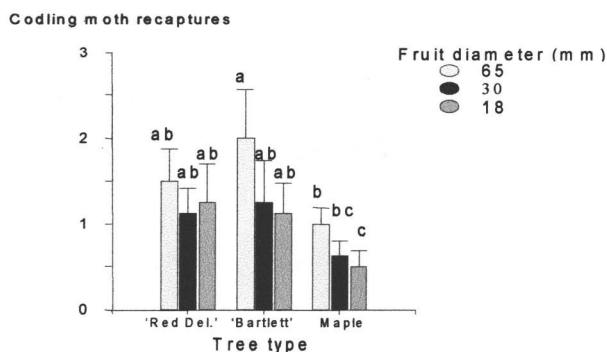


Fig. 3.10: Codling moth adult recaptures on trees with fruit of different size. Each value is the combined male and female mean over the three-day period after release for each tree/fruit type combination. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Fruit shape (apple or pear) was not a factor orienting codling moth adults ($F = 0.61$; $df = 1, 3$; $P = 0.493$). Color appeared to be an important factor ($F = 11.76$; $df = 2, 6$; $P = 0.008$), being trees with green or yellow fruits favored as host over trees with red fruits (Fig. 3.11). Same results were obtained on trees with fruit of both shapes ($F = 0.40$; $df = 2, 6$; $P = 0.686$, for ANOVA testing significance of interaction factor between fruit color and shape).

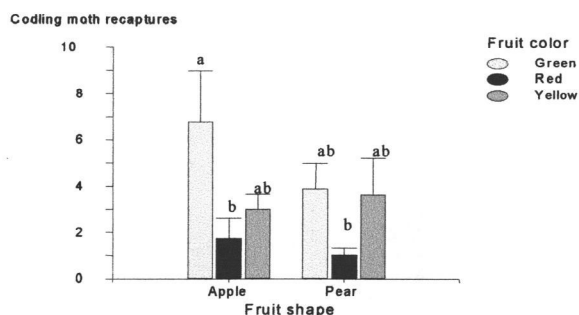


Fig. 3.11: Codling moth adult recaptures on trees with fruit of different shape and color. Each value is the combined male and female mean over the three-day period after release for each color/shape fruit combination. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Effect of fruit density on host selection. Fruit density seemed to be definitely a factor for host selection ($F = 9.22$; $df = 3, 32$; $P = 0.0002$). There was a positive relation, but not linear, among fruit density and moth catches (Fig. 3.12).

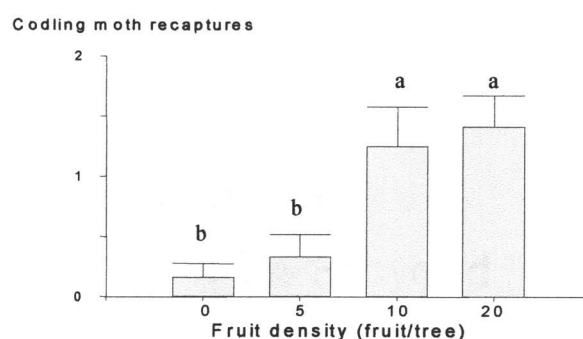


Fig. 3.12: Codling moth adult recaptures on trees with different fruit density. Each value is the combined male and female mean over the three-day period after release for each fruit density level. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Host selection behavior of different codling moth populations. Adults of both codling moth populations showed a similar behavior pattern during host selection, but the statistical analysis showed significance of the interaction among strain x fruit type ($F = 18.64$; $df = 2, 6$; $P = 0.002$). Interaction among strain x tree type approached significance ($F = 3.63$; $df = 2, 6$; $P = 0.093$). For individuals of the Canadian population both tree and fruit type were key factors controlling codling moth adult response in host selection (Fig. 3.13). Individuals of the MCAREC population preferred trees with natural fruit as host over trees with artificial fruit and with no fruits; moths were more attracted to trees with artificial fruit than to trees with no fruit, but this different preference was not significant (Fig. 3.13). Adults were significantly more attracted to 'Red Delicious' trees, but they did not discriminate among host 'Bartlett' trees and non-host maple trees.

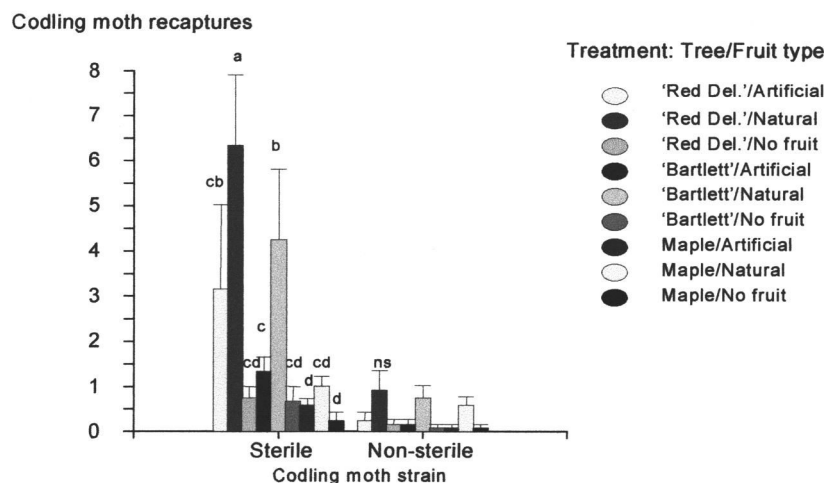


Fig. 3.13: Recaptures of adults of a sterile and a non-sterile codling moth strain when exposed at different tree types with different fruit types as a potential host. Each value is the combined male and female mean over the three-day period after release for each tree/fruit type combination. For each strain, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

3.4 Discussion

Moth recapture efficiency of interception traps was very low (1.91% and 8.75% of the sterile Canadian and non-sterile MCAREC populations, respectively) compared to the recaptures obtained by Weissling & Knight (1994), who reported recapture rates above 20% in experiments with non-sterilized laboratory reared moths. Higher recapture rates obtained by these authors could be related to better weather conditions for codling moth flight the days they conducted the experiments. However, these authors did not support their results with weather data. No adults were recaptured when hourly average temperature during the period of flight activity was lower than 12 °C. This is associated to the fact moths do not fly in those conditions. These observations agree with lower temperature thresholds for flight activity reported in the literature that range from 11 to 16°C (Zech 1955, Riedl et al. 1986). Song & Riedl (1985) reported that the proportion of flight-active males increases with temperature according to a cumulative normal distribution. They reported that male

moths begin to fly at 11 °C, 50 percent are active at 14 °C, and the optimum temperature range for male flight is between 18° and 26 °C in laboratory experiments. Although rain inhibited codling moth flight during the experiments and was a big factor decreasing codling moth recapture, it was too sporadic to explain the low recapture rates during the whole season. Wind was not a critical factor for codling moth flight. Wind speeds up to 10 km/h did not inhibit codling moth flight. However, we recorded data of hourly average and maximum wind speeds. Instantaneous wind speeds would be more useful to understand the effect of wind on codling moth flight. During an hour period wind conditions can fluctuate significantly in the experimental sites. Even though maximum and average hourly wind speed reached high values there could be a time interval with no wind or low wind speed long enough during an hour period to allow codling moth flight. Pristavko (1969) reported that wind speeds of 1 m/sec (=3.6 km/h) inhibit codling moth flight activity. Pre-release conditioning of the moths could also have played a role decreasing recapture rates. Petri dishes (9 cm diameter) were used to store 800 adult moths of the sterile population. The performance (flight capacity) of the sterile moths could have been affected by the fact there was a longer time interval (from a few hours to 3-4 days) between emergence and release compared to the experiments conducted by Weissling & Knight (1994). The high release density (270 moths/cup at the MCAREC, and 800 moths/cup at The Dalles site) could be another probable explanation.

Significantly more males were recaptured in the experiments. These observations may suggest that females are more sedentary than males.

The analysis of recapture data suggested that codling moth adults do not orient to and select host plants at random, but they have a directed movement in response to cues from host plants. The relative importance of visual and olfactory stimuli from canopy and fruit as cues for codling moth host selection varied through the season. Stimuli from the canopy seemed to play an important role during the whole season. Preference for pear over apple by codling moth adults in the experimental site at the MCAREC did not correspond with Westigard's et al. (1976) findings that apple is more susceptible to codling moth than either 'Bartlett' or 'Anjou' pears. Observations suggest that host preferences by adult moths do not explain differences in susceptibility to codling moth attack among certain apple and pear

cultivars. The consistently low number of moths caught in maple trees compared with host trees suggests that codling moth adults follow olfactory and/or visual cues when seeking out hosts. It is possible that host plants (apple and pear) release volatiles that act as attractants and/or as arrestants. Non-host maple trees, on the other hand, either lack volatiles acting as attractants/arrestants or these compounds are released in quantities too small to be detected by codling moth adults and used as cues for locating them as host. Moths did not avoid maple trees altogether as evidenced by low moth catches on these non-host trees. This suggests that maple foliage is not repellent but at the same time it may not be attractive either. The low catches in the maple trees may merely be a result of random movement of moths and are likely not a directed response to certain attractive cues. However, fewer recaptures in non-host maple trees could be partially related to a smaller canopy volume in our experiments at the MCAREC site.

The role of fruit stimuli on the host selection response of moths varied through the season but was consistent among cultivars. Early in the season, when the fruit is still small (diameter 18 mm), fruit seems not to have any effect on adult moths response on host selection. At that time of the year, fruit is probably too small to be perceived visually by adult moths at the distance they were released from potential hosts. The amount of volatiles released by natural fruit that affect adult behavior might also be too small and be masked by stimuli from the canopy. The data suggest that at the beginning of the season cues associated with the canopy or foliage are primarily responsible for guiding codling moth adults to host trees. However, the significance preference for trees with natural and artificial fruit over trees with no fruit during the rest of the season suggest that visual and/or olfactory cues associated with fruit enhance the response of codling moth adults to host. Trees with natural fruit (either infested or non-infested) were especially favored as host over trees with artificial fruit late in the season. This suggests that in late summer olfactory cues may be more important than visual cues for attracting moths to host trees. However, Wildbolz (1958) proposed that codling moth females were guided primarily by visual cues and that movement of codling moth females between closely planted trees in a uniform orchard was random.

Moths showed a similar preference for trees with natural non-infested and natural codling moth infested fruit during the whole season. This suggests that quality and quantity of chemicals released by infested fruit did not have antixenotic effects on moths during host selection.

The relative importance of cues associated with foliage and fruit was consistent among cultivars. However, results of the interactions among tree and fruit type suggest that the relative roles of cues from tree and fruit type may not be the same in all cultivars. For the 'Anjou' cultivar cues associated with foliage seemed to play a greater role than those from fruit through most of the season in attracting moths. However, this was not the case with 'Newtown' and 'Bartlett' trees.

At the end of the season, trees of the 'Bartlett' cultivar with natural non-infested and infested ripe fruit were equally favored as hosts and preferred over trees with artificial fruit. This suggests that volatiles associated with fruit maturity, such as the ripening agent ethylene, do not have antixenotic effects on moths during host selection.

Results from the experiment conducted at the MCAREC site demonstrated that fruit adds to the overall attraction of a host tree once it reaches a certain state of maturity. The results also suggested that importance of stimuli from canopy and fruit depends on factors such as the relative surface area of leaves and fruits, and consequently on the canopy volume and the density and size of fruits, as well as on the plant species and cultivar. In an experiment at The Dalles site, artificial trees with natural fruit were not significantly preferred over trees with natural foliage and artificial fruit. The leaf to surface ratio was 10:1 approximately. This suggests that, in terms of amount of attractants released per unit of surface area, olfactory stimuli from fruit are more important than olfactory stimuli from the canopy for codling moth responses on host selection.

Codling moths released at 5, 10, and 15 m from potential hosts responded in a similar way. However, the results suggest that the relative importance of visual and olfactory stimuli both from the canopy and the fruit guiding codling moth adults to a plant may

depend on the distance to the source where moths detect the stimuli. At short distances both olfactory stimuli from foliage and visual and olfactory cues from fruit seem to be key factors on host selection. At longer distances, olfactory stimuli from canopy and visual factors from fruit seem to have a less important role, and moths are mostly guided by olfactory stimuli from host fruit. Stimuli from canopy would probably play a more important role guiding adult moths from longer distances if trees had a higher leaf /fruit surface ratio. Visual cues from fruit would also be more important if fruit size was bigger. The fact that the distance between trees increased with the distance from the release point could also help discriminating stimuli from different choices at longer distances.

Alpha-farnesene is a volatile released by pome fruits (apple, pear) and reportedly affects oviposition behavior of codling moth females (Sutherland et al. 1974) and searching behavior of neonate larvae (Landolt et al. 1998). However, results showed no correlation between the preferences moths displayed for the two pear cultivars 'Anjou' and 'Bartlett' in the release experiments and the α -farnesene content in leaves and fruit. This finding does not exclude α -farnesene as a potentially attractive agent in host selection since α -farnesene may be active at amounts below those found in 'Anjou' pears. Moreover, it would be important to know how α -farnesene behaves in the environment once released by host fruit and leaves. Anet (1969) reported that α -farnesene rapidly oxidizes to conjugated triene hydroperoxides. Therefore, it is likely that α -farnesene does not have any effect at long distances from its source. This would imply that its role on adult host selection is very limited. Other chemical factors may play a role in host selection but were not investigated here. Bäckman (1999) reported that female moths responded to several volatiles from apple branches with leaves and fruitlets other than (*Z,E*)- α -farnesene and (*E,E*)- α -farnesene. These included 4,8-dimethyl-1,3(*E*), 7-nonatriene, linalool, β -caryophyllene, β -farnesene, germacrene D, and methyl salicylate. Attraction to a host tree could involve one or more of these compounds. Similar studies with different pear cultivars are necessary to understand the biological significance of plant volatiles and what role they might play in host selection.

Codling moth adults may also be able to discriminate among host trees because of differences in visual characteristics of foliage and fruit. Results suggest that visual cues

from canopy and fruit shape do not greatly affect adult response on host selection. Dimensions of canopy are probably also important guiding codling moth adults, but this should be confirmed. Wildbolz (1958) recaptured more moths in groups of apple trees than in individual apple, pear, and non-host trees. Foliage of the different plant species and cultivars used in the experiments had similar reflectance patterns and color attributes; artificial fruit had also similar reflectance curves and color attributes than natural fruit, suggesting that visual factors of artificial material did not affect the response of adult moths compared to visual factors of natural plant material. However, results show that fruit size is a visual cue perceived by codling moth adults. Moreover, adult moths showed preference for green and yellow artificial fruit over red fruit. Since 'Red Delicious' trees (red fruit) were at least as preferred as 'Bartlett' trees (green or yellow fruit), this finding seems to support the hypothesis that olfactory stimuli from plants play a more important role than visual cues on host selection by the adult stage. Other insects, including various fruit flies, such as the apple maggot, *Rhagoletis pomonella* (Walsh) (Still 1960, Oatman 1964, Prokopy 1968, 1972), the cherry fruit fly, *Rhagoletis cingulata* (Loew) (Frick et al. 1954), and the walnut husk fly, *Rhagoletis completa* Cresson (Riedl & Hislop 1985) have a more directed response to visual cues.

Fruit density seems to be definitely a factor on host selection, being a positive relation, but not linear, among fruit density and moth catches. Olfactory, as well visual, cues of the fruit are likely involved in this positive correlation between adult response and fruit density. Suski et al. (1981) related fewer adult males within the release orchard to low fruit sets.

In the experiment comparing the response of individuals from two populations, a similar behavior pattern during host selection was observed. However, for sterile individuals of the Canadian population both tree and fruit type seem to be key factors controlling their response on host selection. Individuals of the MCAREC population rely more on olfactory cues from natural fruit to select the host plant. The role of stimuli from the canopy seems to be less important. These results could be related to the fact that this population is reared with natural apples and its individuals might keep in their "memory" more about olfactory cues that guide them to the host tree. Following this hypothesis, a wild strain would be expected to behave more similarly to the MCAREC than to the Canada

population. The fact we worked with a lower number of adults of the MCAREC population also could have led to find these differences.

These observations suggest that sterile moths, in spite of years of laboratory rearing and radiation treatment with Cobalt-60, behave similarly to non-sterile moths reared on apples and still respond to and recognize host trees. This is important, since the fruit industry in British Columbia, Canada, is presently engaged in a multi-year sterile insect release (SIR) program to control codling moth in a limited geographic area.

3.5 Conclusions

Codling moth adults do not orient to and select host plants at random. They have a directed movement in response to cues from host plants during host selection.

There is no correlation between adult preference as host and codling moth infestation level observed in the field among certain host cultivars. Therefore, differences in susceptibility to codling moth damage among apple and pear cultivars are not likely a manifestation of host preference by the moth stage. Maple trees are less preferred than apple and pear trees, but not avoided as host. This suggests that non-host trees do not release volatiles that act as repellents. However, they probably do not produce attractants and/or arrestants in enough amounts to trigger a positive response in adult moths. This could partially explain the lack of success of the codling moth colonizing and developing on non-host species.

Adult codling moths rely on olfactory stimuli from canopy to select a host during the whole season. Both visual and olfactory stimuli from host fruit are important to guide adult moths to a host tree, except at the beginning of the season. Small fruit might not be detected visually from the distances moths were released from potential hosts. Moreover, at the beginning of the season olfactory stimuli from small fruit can be masked by olfactory cues from the canopy. Olfactory stimuli from fruit seem to be more important than olfactory cues from canopy in terms of amount of volatiles released by unit of surface area. Moreover, olfactory stimuli from host fruit seem to affect adult orientation in a wider range than both olfactory stimuli from canopy and visual stimuli from fruit. Volatiles released by

ripe 'Bartlett' fruit, such as ethylene, and infested fruit do not seem to have antixenotic effects on moths during host selection.

Size (bigger fruit is more attractive) and color (red color is less attractive relative to yellow and green) of the fruit affect moth response on host selection, but fruit and canopy shape do not. Attraction of moths to trees increases as host fruit density increases. Differences in preference as host among host cultivars and trees with different fruit density could be used as a management practice. Trees of a more preferred cultivar that have high fruit density could be interplanted in orchards of conveniently thinned trees of less preferred cultivars. Hence, trees of the preferred cultivar would act as an attractive trap where a selective control measure could be applied.

Non-sterilized individuals reared on natural apple relied more on olfactory cues from fruit to orient to a host plant than sterilized moths reared on diet. However, a similar behavior pattern was observed in different strains. Furthermore, the differences observed could be statistically related to the fact that few releases were made with non-sterilized moths. This suggests the control tactic based on the release of sterile moths released in commercial apple and pear orchards may be efficient. Since sterile moths are attracted by cues from host trees they will tend to stay in the orchards, especially if fruit is available. This increases the probability of mating with feral moths and, consequently, it leads to a better control of the pest.

CHAPTER 4
CODLING MOTH (LEPIDOPTERA: TORTRICIDAE)
MATING AND OVIPOSITION BEHAVIOR
ON APPLE AND PEAR CULTIVARS

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4.1 Introduction

The codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), attacks pome and stone fruits as well as walnuts. Big differences on codling moth damage exist among host species. It is most destructive on apple and pear. There are also big differences among apple cultivars (Felt 1910, Newcomer & Yothers 1924, Cutright & Morrison 1935, Witehead 1944), but especially on pear (Westigard et al. 1976). Differential mating and oviposition level, as well as oviposition behavior leading to a differential egg distribution, may be factors explaining differences in infestation levels among host species and cultivars.

Castroville & Cardé (1979) determined that, like the other reproductive behaviors in the codling moth, mating followed a circadian rhythm. Mating takes place during late evening or after sunset depending on temperature. Several authors (Hutt & White 1977, Castroville & Cardé 1979, Castroville & Cardé 1980) have shown the role of visual and olfactory stimuli from the female on the mating process. Whether mating can occur on a non-host tree is unknown. Moreover, almost no information exists about the effect of stimuli from host plants to trigger calling behavior on female codling moths.

Selection of an appropriate oviposition site is the next step for a mated female. Very few eggs are laid on distant parts of a tree away from fruit (MacLellan 1962, Geier 1963, Wearing et al. 1973). These observations suggest that the search for a suitable oviposition site may be directed as opposed to random and that certain cues guide a female to a fruit cluster. Visual (shape and color), chemical/olfactory and tactile stimuli from fruit may be involved in the selection of an oviposition site. These stimuli may directly influence the development of ovaries and the number of eggs actually deposited (Deseö 1970). Wildbolz (1958) has shown that fruit odors attract a female to an oviposition site. Wearing & Hutchins (1973) and Gut (1980) suggested that alpha-farnesene, a naturally occurring sesquiterpene produced by apple, pear, and quince fruits, could act as an oviposition stimulant and attract female moths to a potential oviposition site. The same authors suggested that cultivar susceptibility could be related to differences in the quality and

quantity of fruit odors. Lack of fruit in a host tree could cause a female to disperse until a tree with fruit is located. Various authors (Geier 1963, Wood 1965, Jackson 1979) studied the egg distribution in apple cultivars, but no agreement exists whether the distribution of eggs between clusters is random with regard to individual fruits or clusters. Several studies, both on apple (Hall 1929, VanLeeuwen 1939, Putman 1963, Geier 1963, Wood 1965, Jackson 1979, Subinprasert & Svensson 1988, Blomefield et al. 1997) and pear (Hatting 1942, Hatting 1943, Westigard et al. 1976), have been conducted to investigate the within-cluster egg distribution of the codling moth. Most codling moth eggs are laid on leaves near fruits but a few are laid on the fruit. The proportion of eggs laid on different parts of a fruit cluster varies with host species and cultivar and through the season (Hatting 1943, Putman 1963, Geier 1963, Westigard et al. 1976, Blomefield et al. 1997). Physical characteristics of the surface such as pubescence or presence of wax scales have also been shown to affect egg-laying behavior. Several authors have observed that adult females avoid egg laying on pubescent surfaces on apple (Hall 1929, VanLeeuwen 1939, MacLellan 1962, Putman 1963, Geier 1963, Wood 1965, Hagley et al. 1980), walnut (Olson 1977) and peach (Curtis et al. 1990, Yokoyama & Miller 1988a). Hagley et al. (1980) suggested that the wax scales on some host surfaces, such as the upper side of pear leaves, could negatively affect egg laying. The selection of a specific plant substrate as a suitable oviposition site in close proximity to the larval food source is crucial. The distance that neonates have to travel to reach a fruit affects larval mortality before fruit penetration (Jackson 1982). Therefore, the location and successful penetration of fruit depends upon both the oviposition behavior of the gravid female and the searching behavior of newly hatched larvae. Since egg distribution within a fruit cluster can be quite cultivar-specific, one can expect that neonate mortality before fruit penetration varies as well between cultivars. This could be one of the reasons why codling moth infestation varies among host cultivars.

The purpose of this study was to assess different questions about the mating and oviposition behaviors of the codling moth that still remain unclear. One main objective was to determine what physical and chemical factors from plants affect mating and oviposition behavior of the codling moth and can explain the egg distribution observed in the field. A secondary goal was to elucidate whether or not the codling moth is able to mate on non-host

trees. The other main goal was to determine the egg distribution in different apple and pear cultivars that are currently cultivated in the Pacific Northwest. This study should add data on codling moth egg distribution and mating and oviposition behavior especially on pear cultivars. This information should be useful to assess the importance of adult mating and oviposition behaviors as potential factors of the different susceptibility to codling moth attack observed among host species and cultivars.

4.2 Materials and Methods

Codling moth populations. A population of *C. pomonella* originally collected in 1991 in an apple block at the Mid-Columbia Agricultural Research and Extension Center (MCAREC), Hood River, OR, was maintained on thinning apples in an insectary at 24 ± 2.5 °C, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h as described by Riedl & Loher (1980). This laboratory population supplied adults for mating and oviposition assays. In addition, during the summer of 1999 moths were collected from infested ‘Bartlett’ pear (Southern Oregon Experiment and Extension Center, Medford, OR) fruits to assess oviposition behavior of feral individuals.

Research sites and plant material. Field sampling was conducted in an apple and in a mixed pear-apple-maple block located at the MCAREC. The apple block was 0.5 ha in size with a tree and row spacing of 6.1 m. It consisted of alternating rows of 31-year-old ‘Red Delicious’ and ‘Newtown’ apple trees. Average height ($H = \text{Mean} \pm \text{SEM}$) and canopy width ($CW = \text{Mean} \pm \text{SEM}$) of the ‘Red Delicious’ trees were: $H = 3.05 \pm 0.24$ m; $CW = 2.55 \pm 0.12$ m. The 0.2-ha pear block consisted originally of alternating rows of 35-year old ‘Anjou’ and ‘Bartlett’ pear trees with a tree and row spacing of 8 m. In 1998, six year-old ‘Newtown’ apple and three year-old maple, *Acer platanifolia* cv. ‘Emerald Queen’, trees were planted in-between pear trees in the tree row. The resultant quadrangular block consisted of 15 trees of each cultivar. Average height ($H = \text{Mean} \pm \text{SEM}$) and canopy width ($CW = \text{Mean} \pm \text{SEM}$) of the different tree species and cultivars in this mixed block were: ‘Anjou’: $H = 3.16 \pm 0.05$ m; $CW = 3.21 \pm 0.11$ m; ‘Bartlett’: $H = 3.13 \pm 0.04$ m; $CW =$

2.85 ± 0.05 m; 'Newtown': $H = 3.15 \pm 0.04$ m; $CW = 2.74 \pm 0.06$ m; maple: $H = 3.11 \pm 0.07$ m; $CW = 2.12 \pm 0.06$ m. Mating experiments were conducted in both blocks. In addition, oviposition site selection tests were carried out under semi-field conditions in large screen cages (1.5 x 1.5 x 2 m high) which were located in a grassy area about 50 m away from the nearest apple and pear trees.

Mating assays. Two experiments were conducted to contrast the hypothesis that codling moth mating is influenced by some aspect of host plants against the hypothesis that it is not.

Experiment 1. The hypothesis was tested that stimuli from foliage and fruit of host plants are essential for codling moth mating. Cylindrical sleeve cages (15 cm diameter, 30 cm long) each with one pair of <1-day-old virgin laboratory moths were placed in the field around small branches of 'Red Delicious' apple and maple trees. Moths were provided with water and 5% sucrose (supplied from cotton wicks) during the experiment and left in the cages for 75 degree-days (lower threshold temperature 10°C). Mating success of pairs placed around leaf clusters with natural non-infested fruit, natural infested fruit, artificial fruit, and with no fruit was compared. 'Red Delicious' apples were used in host and non-host trees. The artificial apples were made of Styrofoam® and matched natural fruit in size, shape and color. Three fruit sizes (mm diameter) were used in experiments: 18 mm (PF3600 040; Wang's International Inc., Memphis, TN), 30 mm (PF5000 040), and 65 mm (PF5006 040). Females were dissected to assess mating status and count spermatophores. In addition, the number of eggs laid on leaves, fruit and woody tissue was also counted. This experiment was repeated three times between May and August 1998. Each test consisted of 10 replications of one adult pair each for each host tree/fruit type combination.

Experiment 2. Additional mating experiments were conducted to test the hypothesis that stimuli from different host fruits may have an effect on codling moth mating. Same procedures as in the experiment above were followed, but tests were done with apple ('Red Delicious'), and pear ('Anjou', 'Bartlett'). Experiments were conducted in May and June 2000. Twenty replications of one moth pair each were carried in each host cultivar. Only natural fruit (ca. 2 cm diameter) was used in the experiment.

Egg distribution in the field. During the summer of 1998, branches of ‘Anjou’ and ‘Bartlett’ pear and ‘Red Delicious’ apple trees were sampled for codling moth eggs in the field to determine the oviposition preference for aspect and height levels, for different host surfaces, and at different times during the season. The sampling unit was fixed and consisted of the first 50 cm from the tip of a branch. Sampling units were chosen with respect to compass direction (N, S, E, and W), height (low: 1.0-1.5 m; high: 2.5-3.0 m) and on the basis of fruit number. Branches with 0, 1, 2, and 3 fruit/cluster were sampled. Sampling was done three times during the season: at the oviposition peak of the first (June 3-12) and second generation (August 3-12), and a third time at the 80% oviposition point of the second generation (around ‘Bartlett’ harvest time: August 30-September 7). The phenology model for the codling moth developed by Brunner & Hoyt (1982) was used to determine the dates of the first and second oviposition peak as well as of 80% oviposition of the 2nd generation. Daily temperature data were obtained from the NOAA (National Oceanic and Atmospheric Administration) weather station at the Mid-Columbia Agricultural Research and Extension Center in Hood River. The Degree-Day Utility (DDU) program (Seaber et al. 1990) was used to calculate the degree-days accumulated from biofix (April 24). The lower and upper development thresholds considered in the model are 10 and 31 °C, respectively. The model uses a single sine curve with horizontal cutoff to calculate degree days accumulated. The estimated date of the oviposition peak of the 1st generation was June 3 (180 degree-days (°C) accumulated from biofix). The estimated date of the oviposition peak of the 2nd generation was August 5 (790 degree-days (°C) accumulated from biofix). The estimated date of 80% oviposition of the 2nd generation was August 26 (910 degree-days (°C) accumulated from biofix). After the oviposition peak of the first codling moth generation, trees were thinned in order to get the same proportion of clusters with different fruit densities. Ten trees were sampled each time which resulted in ten sampling units for each combination of cultivar, compass direction, height level, and fruit cluster density.

Apple and pear trees were also sampled during the oviposition peak of the second generation to compare oviposition and egg distribution on fruit clusters containing infested and non-infested fruit. Branches were selected at random. Only one infested fruit was

present in clusters with infested-fruit. The number of trees and branch units sampled was the same as above.

During 'Bartlett' harvest (80% oviposition of 2nd generation), the number and distribution of eggs laid on branches with mature and immature 'Bartlett' pears were compared. The number of trees and branch units sampled was the same as above.

In 1999, oviposition level and egg distribution was compared on 'Newtown' apple, and 'Anjou' and 'Bartlett' pear trees that had either natural or artificial fruit. Branches with only one fruit were used as sampling units. Ten sampling units were checked on each cultivar. Sampling was done August 16-18. Location of the egg and distance to the nearest fruit were recorded.

Oviposition site selection. In several experiments the hypothesis was tested that search and selection for an oviposition site is a directed response and not a random search. Multiple-choice tests were conducted in which different oviposition surfaces were offered to mated female moths. Tests were conducted during the summer of 1998, 1999, and 2000 in walk-in screen cages in the field. Wooden plywood boxes served as oviposition cages. Dimensions varied depending on the experiment and ranged from 12 x 12 x 12 cm high to 32 x 32 x 18 cm high. Moths were provided with water and a 5% sucrose solution (supplied from cotton wicks) during experiments. The interior walls and bottom of the boxes was lined with pubescent felt cloth to discourage egg laying on surfaces other than the substrates which were tested for oviposition preference. The top of the oviposition cages was made of screen. In preparation for oviposition tests, two pairs of 1-day-old moths were kept in plastic vials for 24 h in the insectary and supplied with a 5% sucrose solution. More than 90% of females were mated after this period (S.M., unpublished data). Two pairs of mated adults were used in each experiment since preliminary studies had shown that at this density moths did not interfere with each other and displayed normal behavior (S.M., unpublished data). Experiments were terminated after 3 days since the majority of eggs are laid during the first 5 days after emergence (Riedl & Loher 1980). In order to minimize handling of moths that could affect their performance and behavior, males were not separated from females after the 1-day conditioning period and were present in the oviposition cages during the assays. Most experiments were conducted with laboratory moths but a test was repeated

with field moths collected from 'Bartlett' pear. Plant material for the tests was collected about 90 minutes before use from unsprayed trees in the apple and pear block at the MCAREC. Each experiment consisted of ten replications. Treatments were assigned randomly in all tests and replications. The number of eggs laid on different oviposition substrates was recorded.

Effect of fruit density/cluster and other fruit-related factors on oviposition site selection. Small branches with different fruit treatments were offered to adult moths as oviposition sites in cages measuring 32 x 32 x 18 cm high. Freshly cut branches were immediately placed in a 0.8% solution of Hydraflor® (Floralife Inc., Burr Ridge, IL) to prevent stem plugging and trimmed to a standard 4-5 leaves (plus the growing tip). The base of the small branch extended through the bottom of the cage and was placed in a narrow neck plastic vial with a 5% Floralife® solution (Floralife Inc., Burr Ridge, IL) to keep leaves in optimal condition during the experiment. Twigs were arranged in a square inside the oviposition cages. Distance between adjacent twigs was 20 cm.

The hypothesis was tested that the presence of fruit is important for inducing egg-laying and selection of an oviposition site in experiments with 'Red Delicious' apple and 'Bartlett' pear twigs with different fruit/cluster. Fruit densities in this experiment were 0, 1, 2, and 3 fruit/cluster. Experiments were repeated three times during the season (June 3-6, June 29-July 2, and August 17-20).

In other experiments, the hypothesis was tested that olfactory stimuli from fruit are important for inducing egg-laying and selecting an oviposition site. Experiments were conducted with 'Red Delicious', 'Anjou', 'Bartlett', and maple twigs. Only 'Red Delicious' apples were used in experiments with maple. Egg laying on twigs with natural non-infested, natural codling moth infested fruit, artificial fruit, and no fruit was compared to assess the effect of various fruit characteristics on egg laying. The experiment was conducted three times during the season (May 21-24, June 26-29, and August 6-9).

An additional experiment comparing egg laying on twigs with natural waxed, natural un-waxed, artificial waxed, and artificial un-waxed fruit was conducted with 'Red Delicious' and 'Bartlett' plant material, to determine the role of tactile stimuli on egg laying. The experiment was carried out at the end of May 1999. Natural and artificial fruit were

coated with heated household wax (PAROWAX™, Service Assets Corp., Newport Beach, CA) to create uniform surface characteristics for both.

Oviposition preference on fruits of different host cultivars. In this experiments, the hypothesis was tested that there is an oviposition site preference on fruits of different pear cultivars and apple according to their anatomical and chemical characteristics. Fruits of host cultivars ('Red Delicious', 'Anjou,' and 'Bartlett') were placed at equal distance and presented as oviposition substrate to moths in cages measuring 25 x 25 x 15 cm high. This test was conducted three times during the summer (June 2-4, July 15-17, and August 14-16) to investigate changes in oviposition behavior in relationship to fruit development and ripening. Three fruits of each host cultivar were placed in each replication at the experiment conducted in June. One fruit of each cultivar was used in experiments conducted in July and August.

In a similar multiple-choice experiment, fruits of three different sizes and stages of development (collected May 25, July 10, and August 21 of 1998) were offered as oviposition sites to female moths. The hypothesis was tested that biochemical changes in fruit composition during the process of ripening have some influence on egg-laying and oviposition site selection. The test was conducted in September 1-3 of 1998 with 'Red Delicious', 'Anjou', and 'Bartlett' fruit. Moths were exposed to a similar surface area of fruits of different stages of development. Therefore, the number of fruits of each development stage was different and varied from about 10 fruits collected in May to one fruit collected in August.

Effect of fruit color and shape on oviposition site selection. In this experiment, the hypothesis was tested that visual aspects from the fruit (shape and color) can also be important for inducing egg-laying and selecting an oviposition site. Artificial fruit (3 cm diameter) of different shape (apple, pear) and color (yellow, green, and red) were suspended in oviposition cages (25 x 25 x 15 cm high), to compare the effect of visual characteristics of fruit on oviposition behavior. Color attributes and reflectance curves of the artificial fruits used in the experiment are shown in Table 3.1 and Fig. 3.6 (see Chapter 3). Experiment was conducted in June 23-26.

Oviposition preference on leaf surfaces of different trees. In this experiment, the hypothesis was tested that female moths prefer certain leaf surfaces to oviposit. Moths were given a choice of leaves from three host trees ('Red Delicious' apple, 'Barlett' and 'Anjou' pear) and one non-host (maple). Leaf material of host cultivars was collected from fruit clusters. Leaf disks (2.54 cm diameter) were cut with a gasket punch and placed horizontally on moist paper toweling at the bottom of an oviposition cage (12 x 12 x 12 cm high). Two leaf disks from each cultivar were used in each experiment. A piece of mylar (12 x 12 cm) with 8 circular cutouts was placed over the leaf disks and was covered with felt fabric to discourage egg laying on its surface. The cutouts were 2.3 cm in diameter to expose the leaf disks. Half of the leaf disks of a cultivar had the upper side exposed, the other half the lower side. The test was carried out with laboratory moths three times during the 1998 season (May 28-30, June 26-29, and August 11-13). The experiment was repeated in May 2000 with a field strain from 'Bartlett' pears.

Effect of leaf position and orientation of leaf surface to a fruit and a light source on oviposition site selection. In these experiments, the hypothesis was tested that the position of the leaf and the orientation of the leaf surface to a fruit or a light source have an influence on egg-laying and oviposition site selection. The importance of leaf position (horizontal, vertical) and orientation of the leaf surface relative to a fruit and a light source was studied with 'Red Delicious' and 'Bartlett' foliage. Rectangular pieces (12 x 4.5 cm) of mylar with two circular cut-outs (2.3 cm) were placed over leaf disks (2.54 cm diameter) and placed in various positions (vertical; horizontal with leaf disks facing upwards or downwards) in the oviposition cage. The exposed mylar was covered with pubescent self adhesive wound fabric Fixomull® (Beiersdorf AG, Hamburg, Germany) to prevent egg-laying on surfaces other than plant material. Oviposition cages (12 x 12 x 12 cm high) had screen on the top and bottom to avoid the effect of light on those leaf disks placed horizontally.

To study the effect of leaf position relative to a natural fruit on egg laying behavior, a vertical screen was placed in the oviposition cage to create two sections. Moths and leaf disks were put in one section of the cage while fruit was placed in the other. Leaf disks were suspended vertically in the oviposition cage. A set of two disks was oriented to a fruit while

another set of two disks faced away from the fruit. A single fruit (30 mm diameter) was suspended in the other section of the oviposition cage and located 5 cm from the leaf disks. The experiment was repeated with three other fruit treatments. The first additional fruit treatment consisted of natural fruit that moths could contact through the screen. The second additional fruit treatment consisted of natural fruit that could not be perceived visually. A black screen was placed along with the vertical screen that separated the cage in two sections. This additional screen allowed the moths to perceive olfactory stimuli from fruit. The third additional fruit treatment consisted of artificial fruit that moths could perceive visually. In a similar experiment the fruit was replaced with a 1.5-Watt incandescent light (Ace Hardware Corp., Oak Brook, IL) to study the effect of leaf position relative to the light source on egg laying behavior. The photoperiod during the experiment was 16:8 (L:D) h. The incandescent light was switched on at 5:30 a.m. to coincide with the natural photoperiod at that time of year.

Similar methodology was used to investigate the effect of distance between leaf surface and fruit on oviposition. In this case, leaf disks were placed vertically at 5, 10, and 15 cm from fruit in oviposition cages (25 x 12 x 12 cm high).

Effect of ethylene on mating and egg laying. Two experiments were conducted to test the hypothesis that ethylene affects mating of virgin moths and egg laying of mated moths. Two ethylene concentrations were used in experiments in addition to a control without ethylene (0 ppm): 50 ± 10 ppm, and 100 ± 10 ppm. The 50 ppm concentration is slightly above the average internal ethylene in ripe 'Bartlett' fruits while 100 ppm is around the maximum value. Experiments were conducted in the laboratory at $21 \pm 2^\circ\text{C}$, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h. In the first experiment, one pair of laboratory reared <1-day-old virgin moths was placed inside wax-paper cups (350 ml volume). In the second experiment, one pair of 1-day-old adult moths was placed in the cups. In both cases, adults were provided with water and a 5% sucrose solution (supplied from cotton wicks) and left in the cups for 10 days (about 100 degree-days). Mating success and egg laying of mated females kept under different ethylene concentrations was compared. Female moths were dissected to count the number of spermatophores. Ten replications of one pair of moths each were conducted for each experiment and ethylene concentration.

Analysis of α -farnesene and ethylene. Content of α -farnesene of fresh leaf and fruit of host cultivars ('Red Delicious' apple, 'Anjou' and 'Bartlett' pears) and fresh maple foliage was determined at six different times during the 1998 season. Fruit ranging from 2 to 170 g (10 to 65 mm diameter) and leaves (approximately 10 g per sample) from both fruit and non-fruit clusters were randomly selected for hexane extraction. For each replicate of each treatment, a leaf and fruit sample was individually weighed. Samples were dipped in 100 ml HPLC grade hexane for 3 minutes, turning the fruit and leaves manually during dipping (Meir & Bramlage 1988). Aliquots of the hexane extracts were diluted appropriately and scanned for UV absorption with a spectrophotometer (Model DU-64, Beckman Instruments Inc, Fullerton, CA) between 190 and 300 nm wavelengths. The absorbances were recorded at 2 nm intervals. The mean absorbance of three replicates per treatment was plotted against the corresponding wavelengths at 2 nm intervals to obtain the UV spectrum for each treatment. Alpha-farnesene was measured at O.D. (=absorbancy) 232 ($E_{232} = 27,700$), and was expressed as nmoles/cm² of fruit or leaf.

The internal ethylene concentration on ten mature and immature (infested and non-infested) 'Bartlett' pears at harvest time was determined. The intercellular gas in the fruit was extracted under reduced pressure of 600 mm Hg for 20-25 s (Beyer & Morgan 1970). One ml of extracted gas was immediately injected into the gas chromatograph (Model GC-9A, Shimadzu Corp, Kyoto, Japan) to determine the ethylene concentration.

Measurement of environmental variables. Temperature and relative humidity in sleeve cages, in the walk-in cages, and in the room where ethylene experiments were conducted were recorded with HOBO StowAway loggers (Onset Instruments, Pocasset, MA).

Statistical Analysis. Logistic regression for binomial responses (SAS 1990) was used to compare the level of mating in different host cultivars and non-host species. Analysis of variance (ANOVA) and contrasts for mean comparison according to Waller-Duncan Bayes LSD were carried out with the Number Cruncher Statistical System (Hintze 1997) to compare the level of egg laying on different cultivars and oviposition substrates in

the field and in multiple-choice tests. Data were subjected to the logarithmic [$\log(\text{eggs}+1)$] transformation. Hatched eggs were not included in the analysis of field sampling data. Analyses of field sampling data were supplemented by Chi-square homogeneity tests (SAS 1990) in which the observed distribution of eggs between and within clusters was compared with an expected even distribution of eggs across clusters and oviposition surfaces. Analyses of oviposition site selection experiments were supplemented by Friedman's non-parametric ANOVA since variances were not homogeneous. Untransformed means ($\pm$ SEM) are reported in the tables and figures.

4.3 Results

Mating bioassays. The percentage of adult codling moths which mated under caged conditions was significantly higher in host plants (apple) than in non-host plants (maple) ($\chi^2 = 5.935$; $df = 1$; $P = 0.015$; Fig. 4.1). Moths mated just once during the 75 degree-day period and never more than one spermatophore per mated female was found. In host plants, mating was 100% during the whole season in moths caged with clusters containing natural non-infested fruit, natural infested fruit, artificial fruit and no fruit (Fig. 4.1). However, when host fruit was added in non-host plants, the percentage of mated females increased (Fig. 4.1), although not significantly ($\chi^2 = 0.858$; $df = 3$; $P = 0.836$, for the fruit term; $\chi^2 = 0.703$; $df = 3$; $P = 0.873$, for the species x fruit interaction term). Higher mating activity in non-host plants was observed especially when natural (both non-infested and infested) fruit was added. Mating activity in non-host plants also tended to increase with the size of the fruit as the season advanced. However, this increase was not significant ($\chi^2 = 1.064$; $df = 2$; $P = 0.588$, for the term time; $\chi^2 = 0.707$; $df = 2$; $P = 0.702$, for the interaction term time x species; Fig. 4.1). Average temperature during the time of mating activity (around sunset) was approximately 15 °C (range from 10 to 22 °C), 19 °C (range from 15 to 22 °C), and 24 °C (range from 17 to 29 °C), respectively, for the three times of the season when the test was conducted.

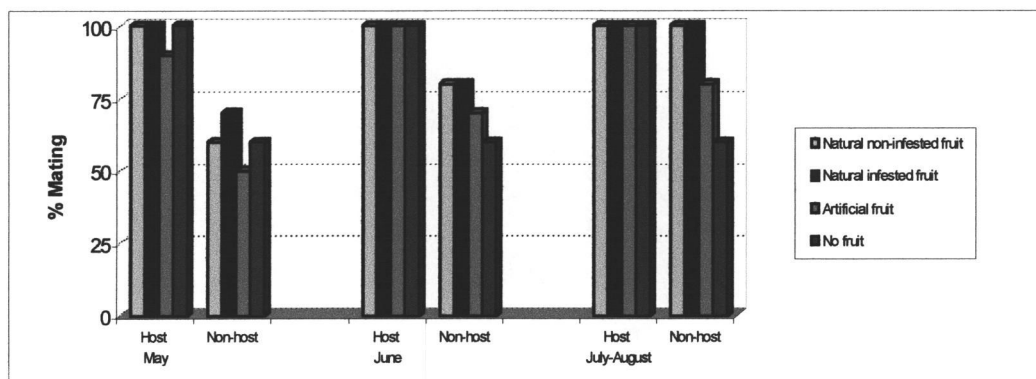


Fig. 4.1: Mating of codling moth in sleeve cages with apple (host) or maple (non-host) foliage with different fruit treatments.

Oviposition activity was significantly higher when mated females were exposed to host fruit ($F = 17.65$; $df = 3, 182$; $P < 0.001$) both in 'Red Delicious' apple and non-host maple trees (Fig. 4.2). No interactions were significant. The addition of artificial fruit to leaf clusters had a positive effect on oviposition activity. However, the total number of eggs laid was higher when natural fruit was added (Fig. 4.2).

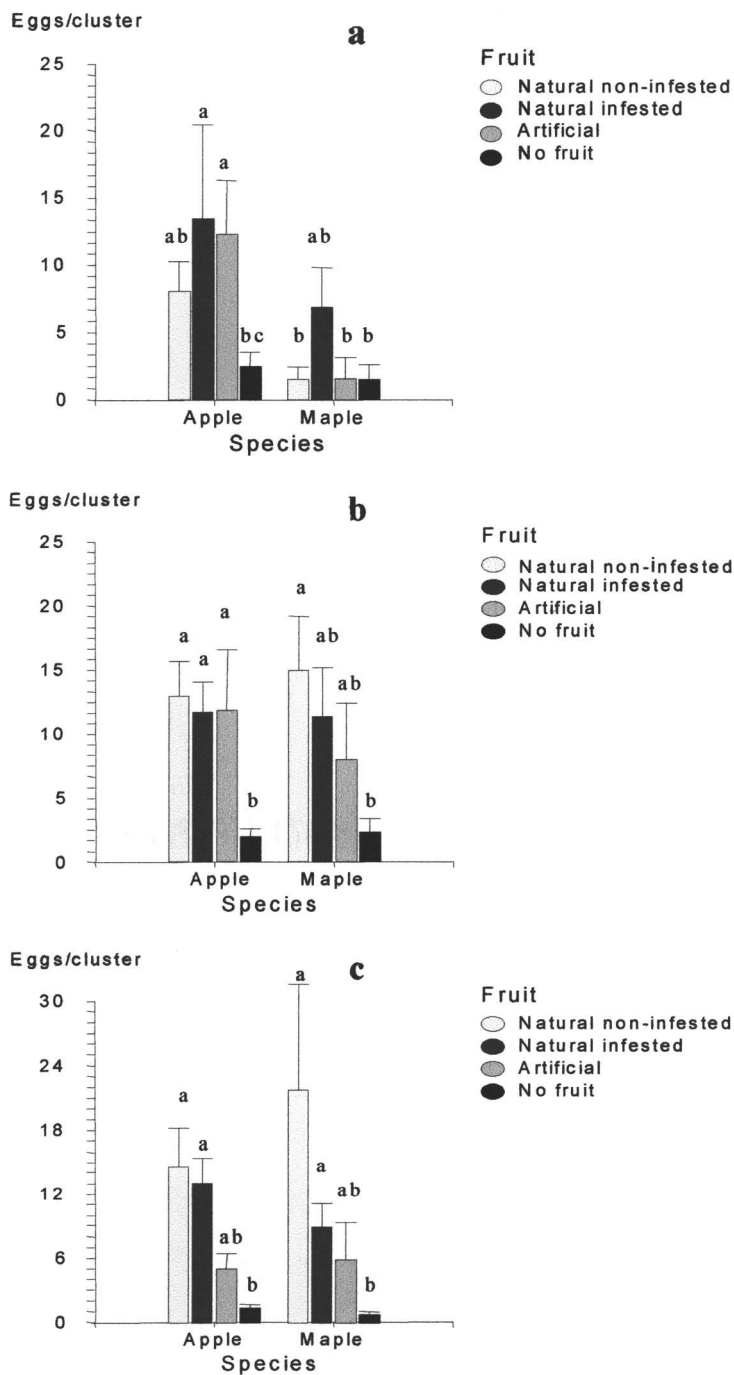


Fig. 4.2: Oviposition of the codling moth in sleeve cages with apple (host) or maple (non-host) foliage with different fruit treatments at three times during the 1998 season: (a) May 10–June 1; (b) June 26–July 2; and (c) August 8–14. For each species and time of season, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

The percentage of adult codling moths which mated under caged conditions was similar in different host cultivars ($\chi^2 = 0.347$; $df = 2$; $P = 0.841$). In different apple and pear host cultivars, mating was practically 100% in moths caged with clusters containing natural non-infested fruit (Fig. 4.3). Average temperature during the time of mating activity (around sunset) was approximately 18 °C (range from 11 to 24 °C) when the experiment was conducted.

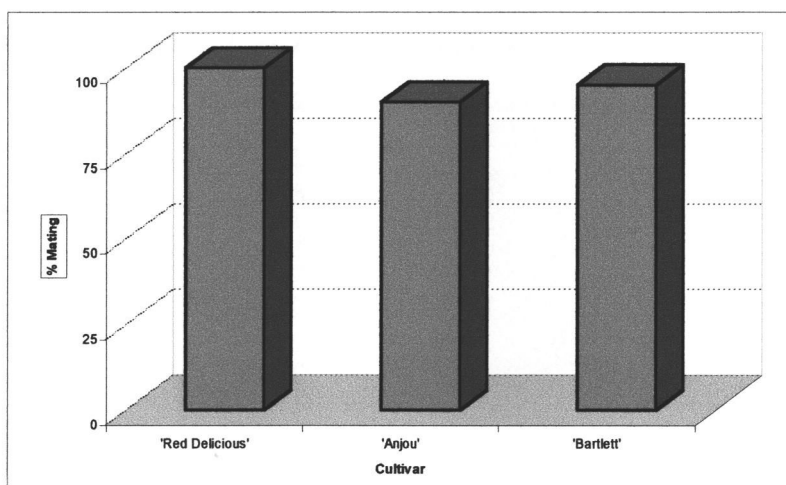


Fig. 4.3: Mating of the codling moth in sleeve cages in trees of different host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear) with natural fruit and foliage.

Egg distribution in the field. Egg laying of field moths at the oviposition peak of the first generation was significantly lower than at the oviposition peak of the second generation ($F = 50.96$; $df = 2, 11363$; $P < 0.001$; Table 4.1). Comparisons of the level of egg laying in different hosts was not appropriate because trees of different cultivars were in different locations. There were no significant differences in the level of egg laying at different tree heights ($F = 3.18$; $df = 1, 11363$; $P = 0.075$).

Table 4.1: Distribution of codling moth eggs in the field on one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars at three time periods during the 1998 season.

Cultivar		1st generation peak	2nd generation peak	80% 2nd generation
'Red Delicious'	Total number of eggs	58	110	58
	% on fruit	37.9	28.2	17.2
	% on upper side of leaf	56.9	52.7	70.7
	% on underside of leaf	5.2	19.1	12.1
	% on wood	0.0	0.0	0.0
'Anjou'	Total number of eggs	39	94	70
	% on fruit	28.2	26.6	61.4
	% on upper side of leaf	28.2	43.6	22.9
	% on underside of leaf	23.1	29.8	14.3
	% on wood	20.5	0.0	1.4
'Bartlett'	Total number of eggs	42	131	61
	% on fruit	38.1	36.6	55.7
	% on upper side of leaf	9.5	9.2	11.5
	% on underside of leaf	40.5	54.2	32.8
	% on wood	11.9	0.0	0.0

Field moths preferred to lay eggs in the east and south over the north and west sections of trees ($F = 4.91$; $df = 3$, 11363; $P = 0.002$). These observations were consistent in all host cultivars ($F = 0.43$; $df = 6$, 11363; $P = 0.859$, for the interaction term orientation x cultivar) during the whole season ($F = 1.25$; $df = 6$, 11363; $P = 0.278$, for the interaction orientation x time).

The within-cluster egg distribution varied through the season ($F = 69.78$; $df = 3$, 11363; $P < 0.001$, for the oviposition surface factor), depending on the host cultivar (Table 4.1). On 'Red Delicious' apple the preferred oviposition surface was the upper side of the leaves through the whole season (52 to 70%). Preference for the fruit surface as oviposition substrate decreased as the season advanced on 'Red Delicious' trees (37 to 17%). On pear,

some eggs were laid directly on the woody surface mostly by moths of the first generation (20 and 12% on 'Anjou' and 'Bartlett', respectively) but never on apple trees. The preferred oviposition surface was the upper side of leaves on 'Anjou' (28 to 43%) and the underside of leaves on 'Bartlett' (40 to 54%) during most of the season. As fruit began to ripen, fruit was the preferred oviposition surface in pear (61 and 55% on 'Anjou' and 'Bartlett', respectively). However, on the 'Bartlett' cultivar (where fruits mature on the tree), female moths did not lay eggs in fruit clusters with yellow ripened fruit (Table 4.2). The within-cluster distribution of eggs on clusters with totally ripened 'Bartlett' pears and immature green 'Bartlett' pears was similar ($\chi^2 < 2.240$; $df = 2$; $P > 0.325$, from homogeneity test in clusters) (Table 4.2). Chemical factors associated to 'Bartlett' fruit ripening may affect oviposition behavior of the moth. The internal ethylene and α -farnesene content of 'Bartlett' pears of different stages of maturity are shown in Table 4.3.

Table 4.2: Distribution of codling moth eggs in the field on the 'Bartlett' cultivar in different size clusters with unripe and ripe pears.

	1 fruit/cluster		2 fruit/cluster		3 fruit/cluster	
	Immature	Mature	Immature	Mature	Immature	Mature
Total number of eggs	14	2	24	3	23	5
% on fruit	57.1	50.0	54.2	33.3	56.5	20.0
% on upper side of leaf	7.2	0.0	12.5	33.3	13.1	20.0
% on underside of leaf	35.7	50.0	33.3	33.4	30.4	60.0
% on wood	0.0	0.0	0.0	0.0	0.0	0.0
Homogeneity test	$\chi^2 = 0.254$; $df = 2$; $P = 0.881$		$\chi^2 = 1.004$; $df = 2$; $P = 0.605$		$\chi^2 = 2.240$; $df = 2$; $P = 0.326$	

Table 4.3: Internal ethylene and alpha-farnesene content in unripe (non-infested and infested) and ripened 'Bartlett' pears.

'Bartlett' fruit (60 mm of diameter)	Internal ethylene (ppm): Mean (SEM)	α -farnesene (nmoles/cm ²): Mean (SEM)
Green unripe non-infested	0.027 (0.001)	0.75 (0.21)
Green unripe infested	0.159 (0.040)	1.49 (0.35)
Yellow ripe non-infested	40.853 (10.116)	1.20 (0.19)

There were significant differences in the number of eggs laid in clusters with different fruit density ($F = 183.99$; $df = 3, 11363$; $P < 0.001$). The number of eggs laid per cluster was correlated with the number of fruit/cluster during most of the season. Twigs with no fruit were not selected as oviposition sites (Table 4.4). Statistical analyses suggest a non-random distribution of eggs among clusters ($\chi^2 > 7.722$; $df = 2$; $P < 0.021$, from homogeneity test with regard to clusters, except on 'Bartlett' trees before harvest time). Eggs were distributed randomly with respect individual fruits ($\chi^2 < 2.215$; $df = 2$; $P > 0.330$, from homogeneity test), except for eggs laid by moths of the first generation. When first generation oviposition peaked, egg laying was concentrated in clusters with 3 fruit/clusters. Fewer eggs were laid on clusters with 3 fruit on 'Bartlett' trees just before harvest time (80% of oviposition of the second generation). There was no difference ($\chi^2 < 10.642$; $P > 0.100$) in the within-cluster distribution of eggs in clusters with different fruit density (1, 2, and 3 fruit/cluster; Table 4.4).

Table 4.4: Distribution of codling moth eggs in the field on one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars in clusters with different fruit density.

Cultivar		1st generation peak	2nd generation peak	80% 2nd generation
'Red Delicious'	Total number of eggs	58	110	58
	% on leaf clusters	0.0	0.9	0.0
	% on 1 fruit clusters	8.9	11.8	12.1
	% on 2 fruit clusters	10.3	31.8	36.2
	% on 3 fruit clusters	82.8	55.5	51.7
	Homogeneity test with respect to fruits	$\chi^2 = 14.020$; $df = 2$; $P = 0.001$	$\chi^2 = 2.1172$; $df = 2$; $P = 0.537$	$\chi^2 = 0.648$; $df = 2$; $P = 0.724$
	Homogeneity test with respect to clusters	$\chi^2 = 28.421$; $df = 2$; $P = 0.001$	$\chi^2 = 18.889$; $df = 2$; $P = 0.001$	$\chi^2 = 7.722$; $df = 2$; $P = 0.021$
	Homogeneity test among clusters	$\chi^2 = 5.730$; $df = 4$; $P = 0.220$	$\chi^2 = 0.835$; $df = 6$; $P = 0.910$	$\chi^2 = 2.441$; $df = 4$; $P = 0.656$
'Anjou'	Total number of eggs	39	94	70
	% on leaf clusters	0.0	0.0	0.0
	% on 1 fruit clusters	0.0	13.8	12.8
	% on 2 fruit clusters	7.7	28.7	32.9
	% on 3 fruit clusters	92.3	57.5	54.3
	Homogeneity test with respect to fruits	$\chi^2 = 16.821$; $df = 2$; $P = 0.001$	$\chi^2 = 1.071$; $df = 2$; $P = 0.585$	$\chi^2 = 0.552$; $df = 2$; $P = 0.759$
	Homogeneity test with respect to clusters	$\chi^2 = 30.046$; $df = 2$; $P = 0.001$	$\chi^2 = 13.401$; $df = 2$; $P = 0.001$	$\chi^2 = 9.392$; $df = 2$; $P = 0.009$
	Homogeneity test among clusters	$\chi^2 = 1.071$; $df = 3$; $P = 0.784$	$\chi^2 = 0.835$; $df = 4$; $P = 0.959$	$\chi^2 = 10.642$; $df = 6$; $P = 0.100$
'Bartlett'	Total number of eggs	42	131	61
	% on leaf clusters	0.0	0.7	0.0
	% on 1 fruit clusters	0.0	17.6	23.0
	% on 2 fruit clusters	7.1	30.5	39.3
	% on 3 fruit clusters	92.9	51.2	37.7
	Homogeneity test with respect to fruits	$\chi^2 = 19.518$; $df = 2$; $P = 0.001$	$\chi^2 = 1.243$; $df = 2$; $P = 0.743$	$\chi^2 = 2.215$; $df = 2$; $P = 0.330$
	Homogeneity test with respect to clusters	$\chi^2 = 32.910$; $df = 2$; $P = 0.001$	$\chi^2 = 12.454$; $df = 2$; $P = 0.006$	$\chi^2 = 1.477$; $df = 2$; $P = 0.478$
	Homogeneity test among clusters	$\chi^2 = 1.425$; $df = 3$; $P = 0.700$	$\chi^2 = 3.222$; $df = 6$; $P = 0.781$	$\chi^2 = 0.398$; $df = 4$; $P = 0.983$

On fruit bearing branches, most of the eggs (> 99%) were laid within 10 cm from the fruit. No significant differences were found in the mean distance of eggs to the fruit among host cultivars ($F = 0.01$; $df = 2, 629$; $P = 0.994$) and among fruit-clusters with different fruit density ($F < 0.01$; $df = 2, 629$; $P = 0.060$).

There was no difference in egg laying in clusters with natural non-infested fruit and natural infested fruit (Table 4.5). The within-cluster egg distribution in clusters with non-infested and infested fruit was similar ($\chi^2 < 5.758$; $df = 2$; $P > 0.056$, from homogeneity test; Table 4.5).

Table 4.5: Distribution of codling moth eggs in the field on one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars in clusters with non-infested and codling moth infested fruit.

Cultivar		1 fruit/cluster		2 fruit/cluster		3 fruit/cluster	
		Non-infested	Infested	Non-infested	Infested	Non-infested	Infested
'Red Delicious'	Total number of eggs	13	15	35	36	61	67
	% on fruit	23.1	40.0	28.6	30.6	29.5	29.8
	% on upper side of leaf	69.2	33.3	42.9	55.6	54.1	46.3
	% on underside of leaf	7.7	26.7	28.5	13.8	16.4	23.9
	% on wood	0.0	0.0	0.0	0.0	0.0	0.0
	Homogeneity test	$\chi^2 = 3.819$; $df = 2$; $P = 0.148$		$\chi^2 = 2.415$; $df = 2$; $P = 0.299$		$\chi^2 = 1.274$; $df = 2$; $P = 0.529$	
'Anjou'	Total number of eggs	13	10	27	25	54	37
	% on fruit	30.8	20.0	29.6	20.0	24.1	27.0
	% on upper side of leaf	46.1	50.0	40.8	44.0	44.4	40.6
	% on underside of leaf	23.1	30.0	29.6	36.0	31.5	32.4
	% on wood	0.0	0.0	0.0	0.0	0.0	0.0
	Homogeneity test	$\chi^2 = 0.373$; $df = 2$; $P = 0.830$		$\chi^2 = 0.675$; $df = 2$; $P = 0.713$		$\chi^2 = 0.160$; $df = 2$; $P = 0.923$	
'Bartlett'	Total number of eggs	23	27	40	40	67	69
	% on fruit	47.8	22.2	40.0	20.0	31.3	18.8
	% on upper side of leaf	8.7	22.2	7.5	22.5	10.5	14.5
	% on underside of leaf	43.5	55.6	52.5	57.5	58.2	66.7
	% on wood	0.0	0.0	0.0	0.0	0.0	0.0
	Homogeneity test	$\chi^2 = 4.177$; $df = 2$; $P = 0.124$		$\chi^2 = 5.758$; $df = 2$; $P = 0.056$		$\chi^2 = 2.959$; $df = 2$; $P = 0.228$	

Egg laying was significantly higher in trees with fruit clusters containing natural fruit than in trees with artificial fruit ($F = 8.97$; $df = 1, 216$; $P = 0.003$). Statistically, the within-cluster egg distribution was not significantly different (homogeneity test: $\chi^2 < 5.324$; $df = 2$; $P > 0.069$) in trees with artificial fruit and in trees with natural fruit clusters. However, egg laying tended to be more concentrated on the foliage in trees with artificial fruit. In trees with natural fruit, eggs were found both on the fruit and foliage (Table 4.6). Eggs were laid farther away from fruit in clusters with artificial fruit (3.3 ± 0.5 cm) than in clusters with natural fruit (1.6 ± 0.3 cm) in all host cultivars ($F = 23.64$; $df = 1, 82$; $P < 0.001$).

Table 4.6: Distribution of codling moth eggs in the field on one apple ('Newtown') and two pear ('Anjou' and 'Bartlett') cultivars in clusters with artificial and natural fruit.

Fruit		'Newtown'	'Anjou'	'Bartlett'
Artificial	Total number of eggs	10	10	10
	% on fruit	10.0	0.0	10.0
	% on upper side of leaf	30.0	50.0	30.0
	% on underside of leaf	70.0	50.0	60.0
	% on wood	0.0	0.0	0.0
Natural	Total number of eggs	21	15	22
	% on fruit	33.3	40.0	31.8
	% on upper side of leaf	23.8	33.3	18.2
	% on underside of leaf	42.9	26.7	50.0
	% on wood	0.0	0.0	0.0
Homogeneity test		$\chi^2 = 1.941$; $df = 2$; $P = 0.379$	$\chi^2 = 5.324$; $df = 2$; $P = 0.069$	$\chi^2 = 1.877$; $df = 2$; $P = 0.391$

Oviposition site selection. *Effect of fruit density/cluster and other fruit-related factors on oviposition site selection.* In tests with 'Red Delicious' and 'Bartlett' branches, there were significant differences in oviposition preference among clusters with different fruit density ($F > 8.56$; $df = 3, 27$; $P < 0.001$). As observed in the field, there was a positive correlation between egg laying and the number of fruit/cluster (Fig. 4.4). Oviposition behavior was similar at the three time periods when the tests were conducted. For each time, average temperature during the time of oviposition activity was approximately 18 °C (range from 15 to 20 °C), 20 °C (range from 17 to 25 °C), and 18 °C (range from 14 to 25 °C), respectively.

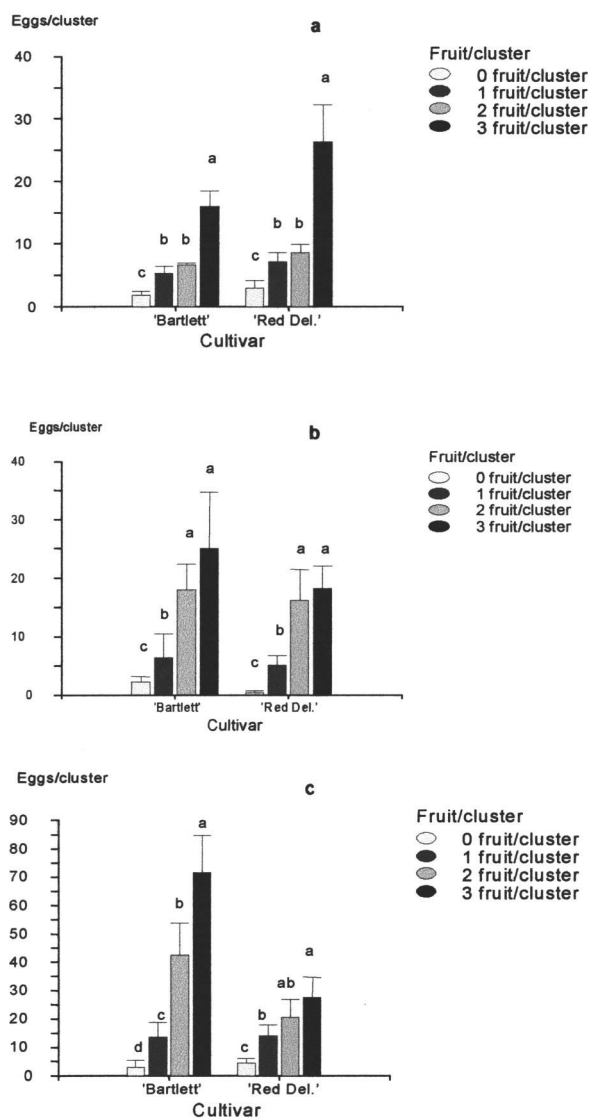


Fig. 4.4: Oviposition of the codling moth in multiple choice experiments on clusters of the 'Red Delicious' and 'Bartlett' cultivars with different fruit density at three times during the 1998 season: (a) June 3-6; (b) June 29-July 2; and (c) August 17-20. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

In multiple-choice experiments, oviposition on branches of different cultivars varied depending on the fruit treatment ($F > 11.62$; $df = 3, 27$; $P < 0.001$). Egg laying was higher on branches with natural fruit regardless of cultivar and time period of season. As under

field conditions with feral moths (Table 4.5), branches with natural non-infested fruit and natural infested fruit were equally preferred (Fig 4.5). Branches with no fruit were clearly avoided. Branches with artificial fruit were significantly less preferred than branches with natural fruit as oviposition sites, especially when fruit was small (May: 18 mm diameter) (Fig. 4.5). Average temperature during the time of oviposition activity (around sunset) was approximately 15 °C (range from 10 to 18 °C), 17 °C (range from 12 to 23 °C), and 22 °C (range from 19 to 28 °C), respectively, for the three times the experiment was conducted.

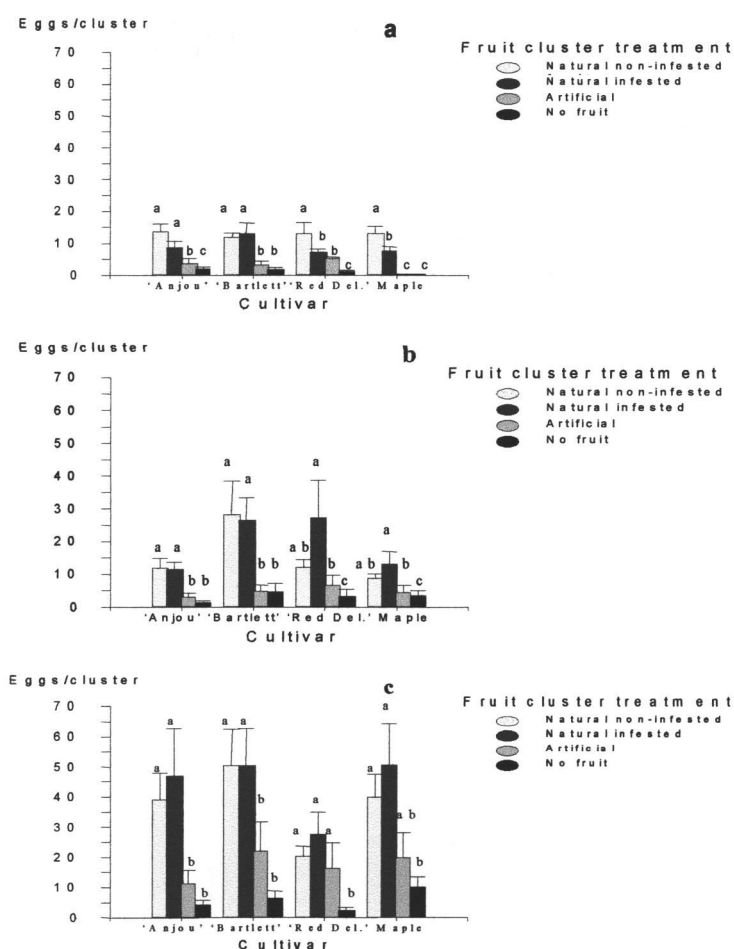


Fig. 4.5: Oviposition of the codling moth in multiple choice experiments on clusters with different fruit treatments (non-infested natural fruit, infested natural fruit, artificial fruit, and with no fruit) at three times during the 1998 season: (a) May 21-24; (b) June 26-29; and (c) August 6-9. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

In another multiple-choice experiment, branches with natural (waxed and un-waxed) fruit were significantly preferred over branches with artificial (waxed and un-waxed) fruit in tests with 'Red Delicious' ($F = 20.39$; $df = 1, 9$; $P = 0.002$) and 'Bartlett' plant material ($F = 68.49$; $df = 1, 9$; $P < 0.001$). Moreover, there was no difference in the oviposition level on waxed and un-waxed artificial fruit in 'Red Delicious' ($F = 1.08$; $df = 1, 9$; $P = 0.326$) and in 'Bartlett' ($F = 0.06$; $df = 1, 9$; $P = 0.819$; Fig. 4.6). Average temperature during the time of oviposition activity (around sunset) was approximately 17 °C (range from 12 to 22 °C) when the experiment was conducted.

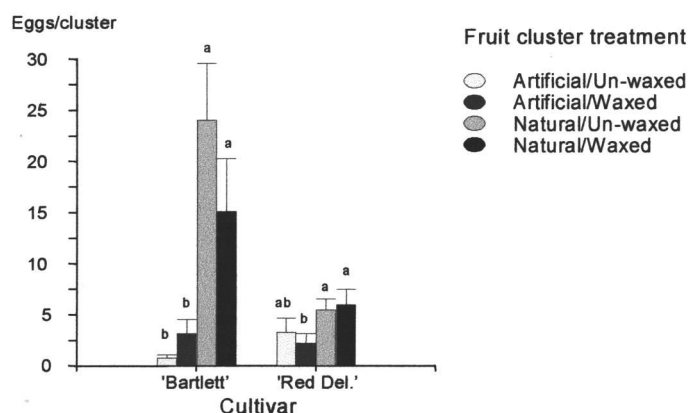


Fig. 4.6: Oviposition of the codling moth in multiple choice experiments on clusters of the 'Red Delicious' and 'Bartlett' cultivars with different fruit treatments (artificial un-waxed fruit, artificial waxed fruit, natural un-waxed fruit, natural waxed fruit). For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Oviposition preference on fruits of different host cultivars. Given a choice, female moths significantly preferred pear fruit to apple fruit as oviposition sites during all three time periods ($F > 6.88$; $df = 2, 18$; $P < 0.006$; Fig. 4.7). Average temperature during the time of oviposition activity (around sunset) was approximately 17 °C (range from 14 to 20 °C), 20 °C (range from 18 to 26 °C), and 22 °C (range from 18 to 26 °C), respectively, for the three times of the season when the experiment was conducted.

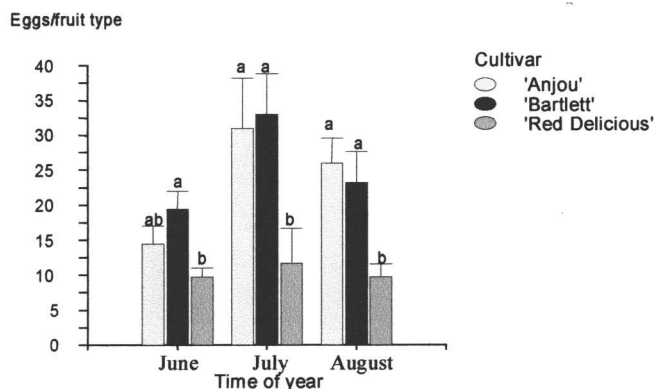


Fig. 4.7: Oviposition of the codling moth in multiple choice experiments on fruit of different host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear) at three times during the 1998 season: (a) June 2-4; (b) July 15-17; and (c) August 14-16. For each time period, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Oviposition on the fruit surface decreased as fruit size increased in 'Red Delicious' ($F = 15.14$; $df = 2, 18$; $P < 0.001$), 'Anjou' ($F = 12.54$; $df = 2, 18$; $P < 0.001$), and 'Bartlett' ($F = 5.96$; $df = 2, 18$; $P = 0.010$) cultivars (Fig. 4.8). Average temperature during the time of oviposition activity (around sunset) was approximately 20 °C (range from 15 to 23 °C), when the experiment was conducted.

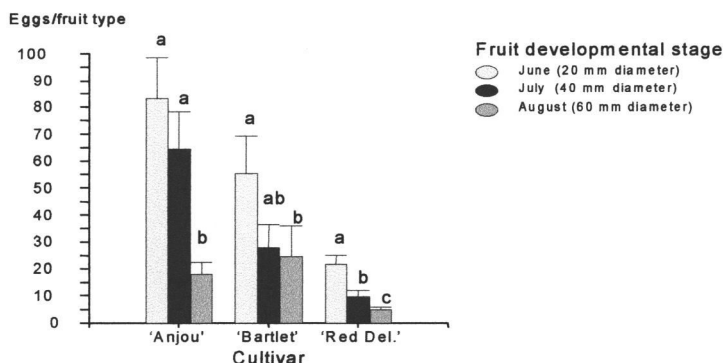


Fig. 4.8: Oviposition of the codling moth in multiple choice experiments on fruit of different stage of development and degrees of maturity of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Effect of fruit color and shape on oviposition site selection. Egg laying was not significantly different on artificial fruits of different shapes (apple and pear) ($F = 0.53$; $df = 1, 9$; $P = 0.486$) and colors (yellow, green, and red) ($F = 1.39$; $df = 2, 18$; $P = 0.274$; Fig. 4.9). However, moths showed the tendency to prefer less both yellow apple and pear fruits as oviposition sites (Fig. 4.9). Average temperature during the time of oviposition activity (around sunset) was approximately 18 °C (range from 13 to 24 °C) when the experiment was conducted.

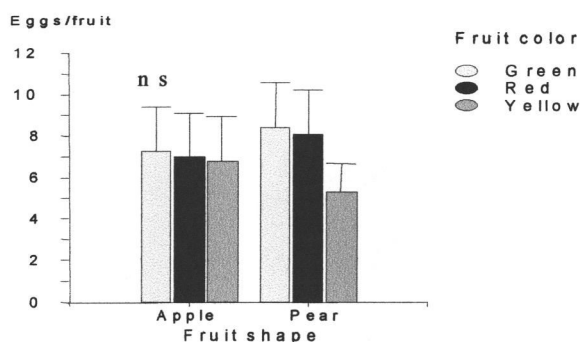


Fig. 4.9: Oviposition of the codling moth in multiple choice experiments on artificial fruit with different shapes (apple and pear) and colors (yellow, green, and red). Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Oviposition preference on leaf surface of different trees. Laboratory female moths laid more eggs on leaf surfaces of host cultivars than on leaves of a non-host species during the whole season ($F > 13.91$; $df = 7, 63$; $P < 0.001$). Females avoided the highly pubescent underside of 'Red Delicious' apple leaves for egg laying. Fewer eggs were laid on both the under and upper side of non-host maple leaves. Oviposition on the non-pubescent leaf surfaces of host plants (upper side of 'Red Delicious', and under and upper side of 'Anjou' and 'Bartlett') was clearly higher (Fig. 4.10a-c). Results were the same for the three time periods when experiments were conducted. Average temperature during the time of oviposition activity (around sunset) was approximately 14 °C (range from 6 to 21 °C), 21 °C (range from 18 to 26 °C), and 21 °C (range from 18 to 26 °C), respectively, for the three times of the season when the experiment was conducted. Field moths collected from infested 'Bartlett' pears showed similar oviposition behavior pattern as moths from the laboratory colony (Fig. 4.10d). However, oviposition preferences for leaf surfaces of pear cultivars were similar to those observed in the field (Table 4.1): moths tended to lay more eggs on the underside than on the upper side of 'Bartlett' leaves and on the upper side than on the underside of 'Anjou' leaves. However, this preference was not significant in the oviposition site selection experiments. Average temperature during the time of oviposition activity (around sunset) was approximately 23 °C (range from 19 to 27 °C) when the experiment was conducted.

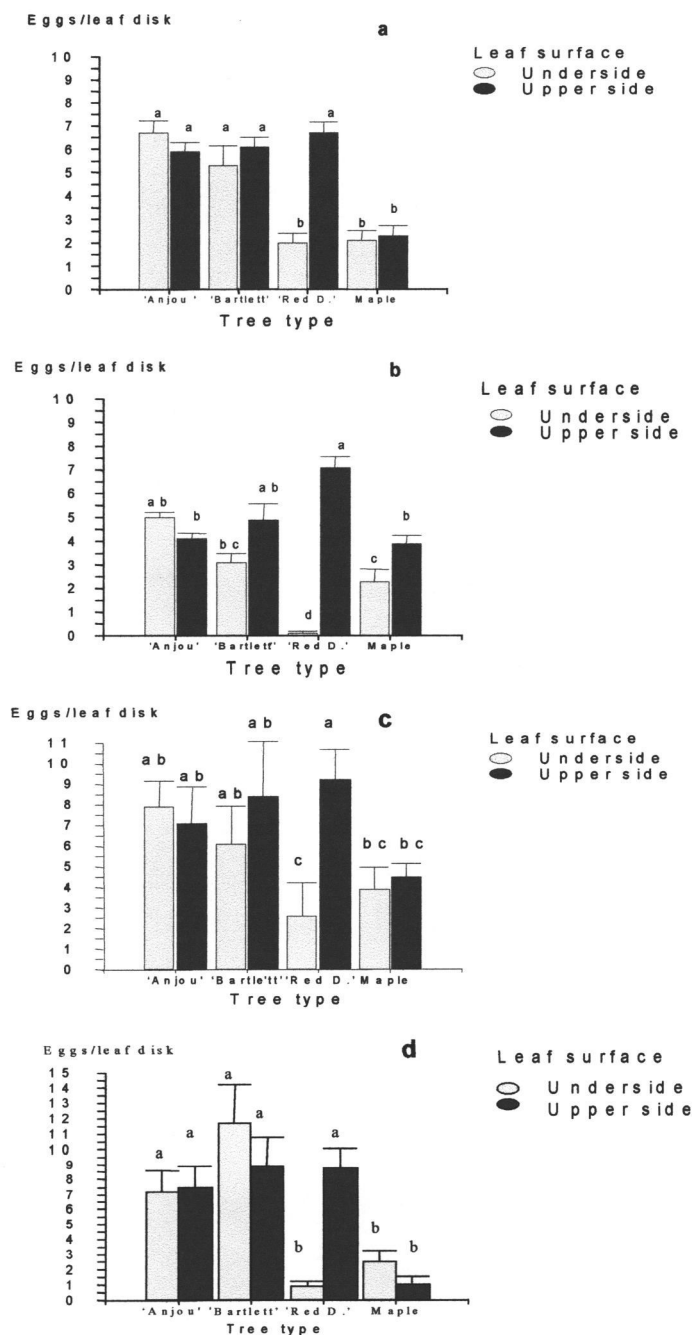


Fig. 4.10: Oviposition of a laboratory codling moth strain in multiple choice experiments on the underside and upper side of leaves of three host cultivars ('Red Delicious', 'Anjou', and 'Bartlett') and one non-host species (maple) at three times during the 1998 season: (a) May 28-30; (b) July 13-15; (c) August 11-13; and (d) a field strain from 'Bartlett' pear. For each time of the season, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Effect of leaf position and orientation of leaf surface to a fruit and a light source on oviposition site selection. Position of the leaf surface was a critical factor in oviposition site selection both in ‘Red Delicious’ ($F = 5.30$; $df = 2, 18$; $P = 0.015$) and ‘Bartlett’ ($F = 12.32$; $df = 2, 18$; $P < 0.001$). Leaf surfaces positioned vertically as well as those positioned horizontally and facing upwards were preferred. Leaf surfaces positioned horizontally and facing downwards were clearly less preferred (Fig. 4.11). The importance of leaf position was greater in ‘Bartlett’. Leaf surface (upper vs. lower side) was not a significant factor in ‘Bartlett’ ($F = 0.02$; $df = 1, 9$; $P = 0.902$; Fig. 4.11b). In ‘Red Delicious’, the hairy underside of apple leaves placed horizontally and facing upwards was less preferred than the upper side of a vertically positioned leaf ($F = 33.18$; $df = 1, 9$; $P < 0.001$; Fig. 4.11a). Average temperature during the time of oviposition activity (around sunset) was approximately 19 °C (range from 14 to 25 °C) when the experiment was conducted.

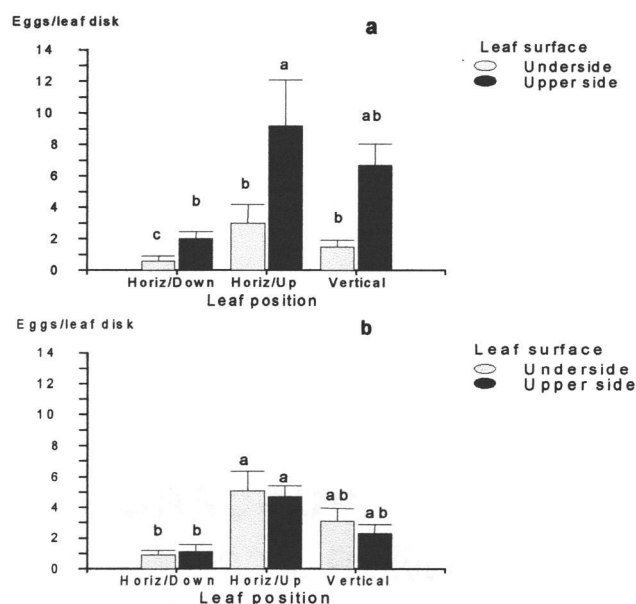


Fig. 4.11: Oviposition of the codling moth in multiple choice experiments on (a) ‘Red Delicious’ and (b) ‘Bartlett’ leaf surfaces placed in different positions (vertical, horizontal). For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Orientation of the leaf surface relative to a visible natural fruit was equally important for oviposition site selection in tests with 'Red Delicious' ($F = 14.74$; $df = 1, 9$; $P = 0.004$) and 'Bartlett' plant material ($F = 18.94$; $df = 1, 9$; $P = 0.002$; Fig. 4.12). Female moths laid consistently more eggs on leaf surfaces facing the light source or the fruit. The role of leaf surface orientation relative to a fruit was relatively more important in 'Bartlett' leaves (Fig. 4.12).

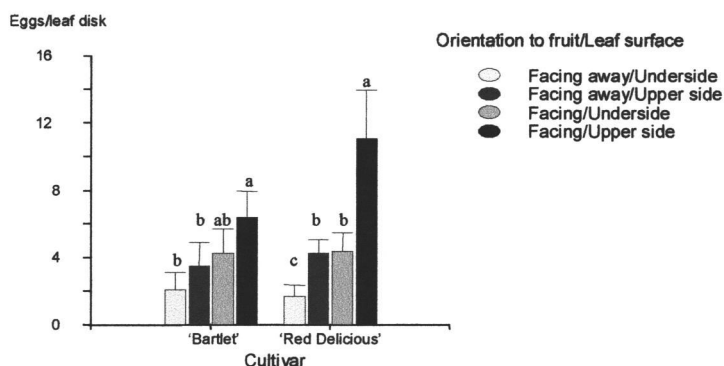


Fig. 4.12: Oviposition of the codling moth in multiple choice experiments on (a) 'Red Delicious' and (b) 'Bartlett' leaf surfaces facing and facing away from a fruit. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

In tests with 'Red Delicious' and 'Bartlett' plant material, female moths laid significantly more eggs on leaf surfaces facing a fruit they could contact but laid fewer eggs on leaf surfaces facing away from the fruit ($F > 5.97$; $df = 1, 9$; $P < 0.03$; Fig. 4.13). Moths also laid significantly more eggs on leaf surfaces facing a fruit they could perceived visually without contact it but laid fewer eggs on leaf surfaces facing away from the fruit ($F > 18.03$; $df = 1, 9$; $P < 0.002$; Fig. 4.13). Oviposition was also higher on leaf surfaces facing a natural fruit hidden from view than on leaf surfaces which faced away from the fruit odor source ($F > 5.81$; $df = 1, 9$; $P < 0.392$; Fig. 4.13). However, oviposition was not significantly different on leaf surfaces facing an artificial fruit and on those facing away

from the fruit ($F < 1.75$; $df = 1, 9$; $P > 0.180$; Fig. 4.13). Average temperature during the time of oviposition activity (around sunset) was approximately 18 °C (range from 13 to 24 °C) when the experiments were conducted.

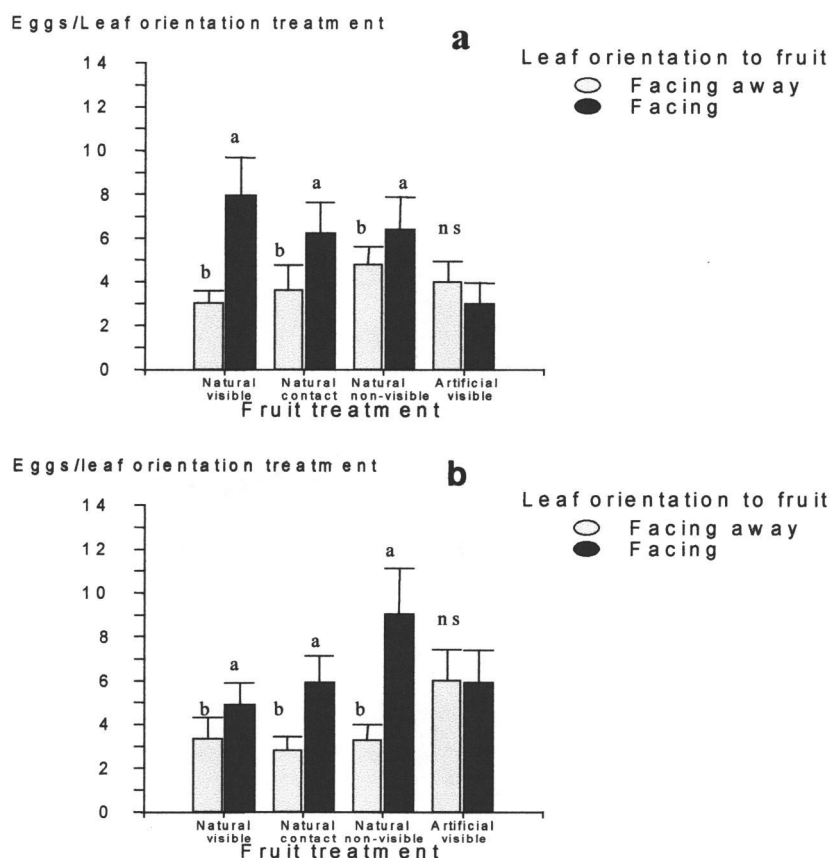


Fig. 4.13: Oviposition of the codling moth in multiple choice experiments on (a) ‘Red Delicious’ and (b) ‘Bartlett’ leaf surfaces facing and facing away from natural and artificial fruit. For each cultivar and fruit treatment, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Orientation of the leaf surface relative to a light source was an important factor for oviposition site selection (Fig. 4.14), both in tests with 'Red Delicious' ($F = 9.24$; $df = 1, 9$; $P = 0.014$) and 'Bartlett' ($F = 18.88$; $df = 1, 9$; $P = 0.002$). Female moths laid consistently more eggs on leaf surfaces facing the light source or the fruit. The role of leaf surface orientation relative to a light source was relatively more important in 'Bartlett' leaves (Fig. 4.14). Average temperature during the time of oviposition activity (around sunset) was approximately 18 °C (range from 14 to 24 °C) when the experiment was conducted.

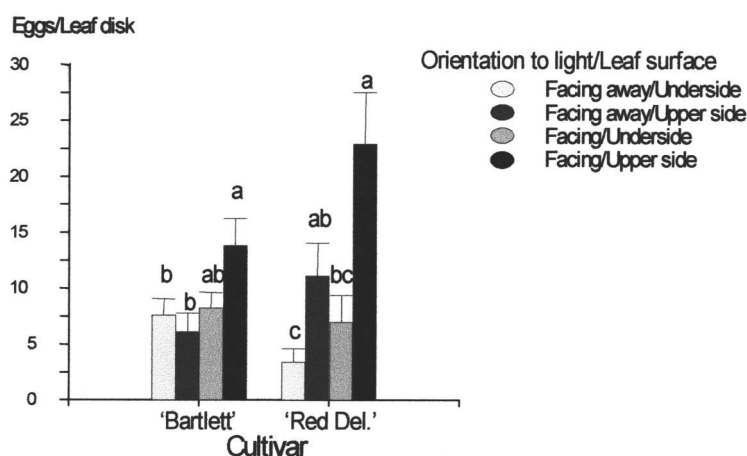


Fig. 4.14: Oviposition of the codling moth in multiple choice experiments on (a) 'Red Delicious' and (b) 'Bartlett' leaf surfaces facing and facing away from a light source. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Oviposition on leaf surfaces varied with the distance of leaves to a fruit in tests with 'Red Delicious' ($F = 49.90$; $df = 2, 18$; $P < 0.001$) and 'Bartlett' ($F > 29.56$; $df = 2, 18$; $P < 0.001$) plant material. Female moths laid more eggs on leaf surfaces oriented to a fruit when leaves were placed at 5 and 10 cm from the fruit. Interaction among 'orientation' and 'distance' was significant ($F > 4.08$; $df = 2, 18$; $P < 0.034$). Orientation to the fruit was not

a significant factor when leaves were placed 15 cm from the fruit and other leaf disks were present at intermediate distances (5 and 10 cm; Fig. 15). Average temperature during the time of oviposition activity (around sunset) was approximately 19 °C (range from 15 to 24 °C) when the experiment was conducted.

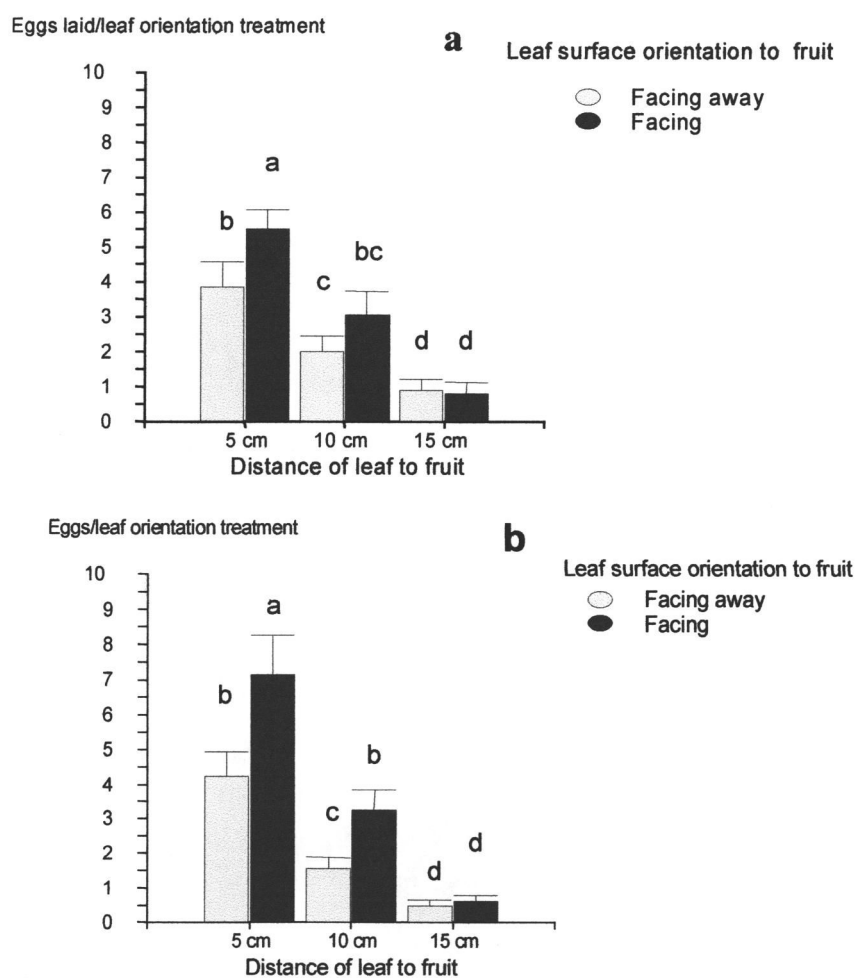


Fig. 4.15: Oviposition of the codling moth in multiple choice experiments on (a) 'Red Delicious' and (b) 'Bartlett' leaf surfaces at different distances from a fruit. For each cultivar, means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Effect of ethylene on mating and egg-laying. Ethylene affected codling moth mating, and may also affect egg-laying (Table 4.7). The percentage of adult codling moths mating under caged conditions was significantly higher without ethylene than with 50 and 100 ppm of ethylene in the air ($\chi^2 = 6.163$; $df = 1$; $P = 0.013$). Egg laying tended to be higher in the absence of ethylene, but differences were not statistically significant ($F = 2.16$; $df = 2, 27$; $P = 0.135$).

Table 4.7: Mating of virgin codling moths and egg laying of mated moths at three ethylene concentrations. Mating rates followed by the same letter are significantly different according to logistic regression for binomial responses. Mean egg layings not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Ethylene concentration (ppm)	n	Oviposition level		Mating (%)
		Mean (SEM)	Range	
0 ppm	10	117.6 (11.7) ns	(39-175)	90.0 a
50 ppm	10	81.1 (14.4)	(9-143)	20.0 b
100 ppm	10	94.4 (10.1)	(41-146)	30.0 b

4.4 Discussion

In host plants, mating was 100% during the whole season in moths caged with clusters containing fruit and no fruit, suggesting that stimuli from host fruit are not essential for mating. Stimuli from host leaves seemed sufficient to trigger mating. However, when host fruit was added in non-host plants, the percentage of mated females increased. These results suggest that although fruit is not essential for mating, it may have a stimulatory role. Mating activity tended to increase in non-host plants with the size of the fruit as the season advanced. Factors from host fruit may be more stimulatory for mating as fruit grows and develops. However, these observations could also be related to the fact that environmental

conditions were closer to optimal for mating as the season advanced. Average temperature during the time of activity around sunset was approximately 15 °C (range from 10 to 22 °C), 19 °C (range from 15 to 22 °C), and 24 °C (range from 17 to 29 °C), respectively, for the three times of the season when the test was conducted.

Oviposition activity was significantly higher when mated females were exposed to artificial and natural host fruit, suggesting that both visual and olfactory cues from fruit have a stimulatory role on oviposition. Several authors have reported that volatiles released by host fruits, such as citronellol (Dethier 1947) and alpha-farnesene (Sutherland 1972, Sutherland & Hutchins 1972, 1973; Wearing & Hutchins 1973, Sutherland et al. 1974, Gut 1980), may act as oviposition stimulants.

Egg laying of field moths at the oviposition peak of the first generation was lower than at the oviposition peak of the second generation. This is likely a result of the inherent difference in fecundity between first and second flight. Gut (1980) related the increase in total oviposition in the field during most part of the season to an increase in the amount of stimulatory fruit volatiles produced as the season progresses and possibly to a similar increase in the amount of foliage available for eggs to be deposited on. The latter would be especially important if the presence of an egg deters further oviposition as was suggested by Wood (1965) and Thiéry et al. (1995).

Field sampling showed that egg laying is similar at different three heights. These observations do not agree with those of several authors who have reported that codling moth oviposition is higher in the upper part of apple trees where mating takes place (Summerland & Steiner 1943, Woodside 1944, Geier 1963, Wood 1965). Blomefield et al. (1997) observed that moths of the first generation preferred to oviposit in the bottom half and second and third generation moths in the top half of 'Golden Delicious' and 'Granny Smith' apple trees.

More eggs were found in the east and south than in the west and north sections of the trees. Although most previous studies did not show a significant effect of aspect on egg distribution, MacLellan (1962) observed that more eggs were laid in the southeast portion of apple trees early but not later in the season. Riedl et al. (1979) reported that pheromone

traps in the north quadrant caught ca. 30% less codling moths than in the other cardinal positions. Therefore, fewer eggs should be expected to be laid in the north quadrant.

The within-cluster egg distribution varied through the season depending on the host cultivar. Our observations agree with those of several authors who have reported the upper side of leaves as the most preferred and the woody portions of branches as the least preferred oviposition surfaces in apple cultivars (VanLeeuwen 1939, Putman 1963, Geier 1963, Wood 1965, Jackson 1979, Gut 1980, Subinprasert & Svensson 1988). However, VanLeeuwen (1939) found that most of the eggs were laid on the underside of the leaves in 'Rome Beauty' apple trees. Blomefield et al. (1997) found that the upper surface of leaves was preferred at the beginning and end of the season and the lower surface of leaves during the middle of the season in 'Granny Smith' and 'Golden Delicious'. A higher percentage of eggs laid on the lower surface of apple leaves could be related to a decreased pubescence later in the season, as suggested by Gut (1980).

The decreasing proportion of eggs laid directly on the apple fruit surface through the season is not in agreement with other authors' observations. Putman (1962) reported that the percentage of eggs laid on the surface of apple fruits increased late in the season. Geier (1963) and Gut (1980) found that the proportion of eggs laid on fruit in apple trees was low in the spring, rose in mid-summer, and decreased gradually towards autumn. Wood (1965) found no seasonal change in the distribution of eggs on apples.

Hattingh (1942, 1943) found that the underside of the leaves was the preferred oviposition surface in the majority of pear cultivars sampled, such as 'Bon Chretien', 'Beurre Bosc', and 'Winter Nelis'. In agreement with our observations in pear cultivars, he reported that the number of eggs deposited on the fruit increased through the season, with the fruit being in its most attractive condition for oviposition just prior to normal harvest time. Since more eggs are laid directly on the fruit surface, a higher proportion of neonate larva do not have to search for the food source. Therefore, at this time of the season a lower mortality of first instar larvae while searching for a fruit should be expected in pear cultivars. Hattingh (1943) found that more eggs were laid on the underside of the leaves in the cultivars 'Bon Chretien', 'Beurre Bosc', and 'Winter Nelis', but on the 'Glout Morceau' cultivar slightly more eggs were laid on the upper than on the lower leaf surfaces over the

whole season. He related this cultivar difference in egg distribution to the 'spiral' growth tendency of 'Glout Morceau' leaves. The leaves of this cultivar curled in such a way that similar areas of lower and upper side of the leaf in any one plane were exposed. The leaves of the other three cultivars tended to fold upwards from the mid-rib to expose the lower and close the upper side surface of the leaf. 'Anjou' leaves also have the 'spiral' tendency that could partially explain why more eggs are laid on the upper side of the leaves. Westigard et al. (1976) and Gut (1980) indicated that the favored site for oviposition in 'Bartlett' trees was the underside of the leaves and that the percentage of eggs laid on the fruit did not differ during most of the season and decreased after the normal harvest time. Westigard et al. (1976) found most of the eggs (74%) on the underside of the leaves and fewer eggs on the upper side of the leaf (7%), on wood (8%), and on the fruit (11%) during the whole season. Gut (1980) found 22% of eggs on pear fruits, 28% on upper-leaf surfaces, 44% on lower-leaf surfaces, and 6% on woody portions of pear branches.

As fruit began to ripen, fruit was the preferred oviposition surface on pear. However, attractiveness of natural fruit seems to decrease as fruit size increases. This seems contradictory. Therefore, we suggest that higher level of egg laying on fruit surface of pears when fruit begins to ripen is likely related to a increase of fruit surface relative to foliage surface area. However, moths avoided ripened 'Bartlett' pears for oviposition. This could be related to a chemical factor. Completely ripened 'Bartlett' pears produce 1,500 times more ethylene than immature green 'Bartlett' pears. Gut (1980) also related the decrease in total oviposition and oviposition directly on the fruit surface on ripened fruit to volatiles, such as ethylene, associated with fruit maturity. Laboratory experiments have indicated that ethylene affects negatively codling moth mating and tends to decrease egg laying at the concentrations it is found in the intercellular spaces of 'Bartlett' pears. However, codling moth is exposed to the concentrations in the air outside the fruit. This ethylene concentration and its effect on codling moth mating and oviposition performance should be determined.

Analyses suggest a random distribution of eggs with respect to individual fruits, except for eggs laid by moths of the first generation. At the oviposition peak of the first generation, egg laying was concentrated in clusters with 3 fruit/clusters. This could be

related to the fact that clusters with at least 3 fruit/cluster were present in a higher proportion at the beginning of the season. Fewer eggs were laid on clusters with 3 fruit late in the season in the 'Bartlett' cultivar. One explanation could be that the proportion of clusters with 3 or more fruit was lower in late season due to natural fruit drop. These observations are similar to those of Geier (1963) who indicated that eggs were distributed at random with regard to individual apple fruits. Wood (1965) found that oviposition within clusters of one, two, and three or more fruits per cluster, was random on 'Delicious' but not on 'Gravenstein' apple. On the other hand, Jackson (1979) indicated that the distribution of eggs among apple clusters deviated from randomness and egg laying was concentrated on clusters with a higher number of fruits, although the stimulatory effect on oviposition of individual fruits in a cluster was not completely additive.

The majority of the eggs were found in close proximity to the fruit in the host cultivars sampled. Moreover, mean distance of eggs to the fruit, as well as complexity of branches (number of lateral branching) and travel speed of neonate larvae (Chapter 5), is similar in different hosts. Therefore, even though the egg distribution within a fruit cluster is quite cultivar-specific, one cannot assume that neonate mortality while searching for fruit varies significantly between cultivars. Several authors have indicated that oviposition takes place in close proximity to the larval food source in apple trees. Geier (1963) and Wearing et al. (1973) found that most eggs were laid within 10 cm of a fruit. Similarly, Blomefield et al. (1997) found the majority of eggs within 12 cm of a fruit. MacLellan (1962) rarely found eggs more than 15 cm from fruit.

Egg laying in clusters with natural non-infested fruit and natural infested fruit was similar, suggesting that visual and chemical changes associated with infested fruit do not seem to attract or repel adult codling moth females when searching for an oviposition site. This could be the result of two compensating factors. Infested fruits produce and release higher amounts of α -farnesene (considered a naturally occurring attractant as well as mating and oviposition stimulant for the codling moth) but also higher amounts of ethylene (that affects negatively mating and tends to decrease codling moth egg laying).

The avoidance of clusters with no fruit as oviposition site in multiple-choice tests with different fruit treatments (natural non-infested, natural infested, artificial, and no fruit) suggests that cues associated with the fruit are important for oviposition site selection. Results also suggest that the role of visual factors becomes more important as fruit size increases, since clusters with artificial fruit were selected as oviposition site in experiments with medium and big fruit, but not with small fruit. The fact that artificial fruit did not stimulate oviposition as much as natural fruit could be related to a lack of chemical stimuli (fruit odors), absence of tactile stimuli of artificial fruit surface resulting in avoidance (oviposition deterrent), or a combination of these factors. Another multiple-choice experiment with waxed and un-waxed artificial and natural fruit has shown that avoidance of clusters with artificial fruit as oviposition site is not due to deterrent factors from the artificial surface. Preference for branches with natural fruit in multiple-choice tests with waxed and un-waxed natural and artificial fruit appeared to be related to volatiles acting as female attractants and/or oviposition stimulants. Moreover, on 'Red Delicious', the added wax layer seemed to stimulate oviposition directly on the fruit surface in contrast to the pubescent surface of un-waxed small apples, suggesting that tactile stimuli also play a role in oviposition site selection. On 'Bartlett', the added wax layer seemed to act more as a barrier for releasing volatiles, and oviposition on waxed natural pears was slightly lower than on un-waxed natural pears. Therefore, these results suggest that chemical, visual, and tactile stimuli from natural host fruits are important for oviposition site selection by adult females.

Pear fruit was preferred over apple fruit as oviposition site. In our experiments there was a correlation among α -farnesene content and preference as oviposition surface in host fruits. Gut (1980) found a preference for 'Seckel' and 'Bartlett' pear to 'Gravenstein' and 'Idared' apple cultivars. Russ (1976), Sutherland et al. (1977), and Gut (1980) suggested that oviposition preference for certain cultivars could be correlated to α -farnesene production.

Multiple-choice tests with artificial fruit showed that female moths are not guided by visual characteristics of the color and shape of the fruit to select an oviposition site. These results also sustain the hypothesis that avoidance of ripened (yellow) 'Bartlett' pear fruits after harvest time is likely due to chemical factor(s).

Female moths avoided hairy leaf surfaces of host cultivars and non-hairy leaf surfaces of a non-host species during the whole season. A combination of morphological and chemical factors can explain these results. Several authors have observed that adult females avoid egg laying on pubescent surfaces on apple (Hall 1929, VanLeeuwen 1939, MacLellan 1962, Putman 1963, Geier 1963, Wood 1965, Hagley et al. 1980), walnut (Olson 1977) and peach (Curtis et al. 1990, Yokoyama & Miller 1988a). Hagley et al (1980) suggested that the wax scales on some host surfaces, such as the upper side of pear leaves, could negatively affect egg laying. Lack of oviposition stimulants (such as α -farnesene) or possible release of oviposition deterrents, can explain the lower preference for the smooth leaf surface of non-host maple. These experiments show that chemicals released by host fruits, such as α -farnesene, may act as oviposition stimulants in female codling moths, as suggested by Wearing & Hutchins (1973), Wearing et al. (1973), and Sutherland et al. (1974). However, differences in α -farnesene content in leaf surfaces of different host cultivars are not correlated to female moth preference as oviposition site.

Laboratory and feral moths showed a similar oviposition preference pattern for leaves of different trees. However, preference of feral moths for leaf surfaces of pear cultivars had the same pattern than observed in field sampling. Laboratory moths showed a tendency to lay more eggs on the side of pear leaves that is less preferred by feral moths. This may suggest that laboratory and feral moths oviposition behavior is slightly different.

Other multiple-choice tests showed that not only chemical and anatomical factors of the leaf surfaces are critical for oviposition site selection. The position of the leaf, the orientation of the leaf surface relative to a fruit and a light source, and the distance of the leaf surface to a fruit are also important in the selection of oviposition sites by adult females. Results suggest that preference for leaf surfaces facing or closer to a fruit is primarily related to olfactory cues from the fruit. This could be related to a higher content of α -

farnesene in leaf surfaces facing a fruit, but more analyses are needed to confirm this hypothesis. Leaf surfaces facing a fruit may absorb higher amounts of the α -farnesene released by the fruit compared to leaf surfaces that are not facing a fruit. However, the relative importance of position of leaf and orientation of the leaf surface relative to a fruit and a light source varies depending on the characteristics of the leaf surface. On 'Red Delicious' leaves these factors become less important since both sides of the leaves are quite different anatomically. On 'Bartlett' leaves these factors are more important because both sides of the leaves are more similar anatomically.

4.5 Conclusions

Codling moth mating rate is higher in host plants than in non-host plants. In host plants, stimuli from fruit are not essential for mating. Stimuli from canopy of host cultivars seem to be enough to trigger mating. However, in non-host plants, host fruit has a positive effect both on the percentage of mated females and oviposition activity. Codling moth mating rate is similar in different host cultivars. Therefore, differences in codling moth attack are not due to differences in mating rates in different host cultivars.

Most of the codling moth eggs are laid at distances closer than 10 cm from a fruit in different host cultivars. This suggests that stimuli from the fruit may act as female attractants and oviposition stimulants. The egg distribution varies through the season, depending on the cultivar. However, mean distances of eggs to the nearest fruit are not significantly different among cultivars. Moreover, additional data suggest that the travel speed of neonate larvae when searching for a fruit is similar in different cultivars. Consequently, no important differences in neonate mortality before fruit penetration should be expected among cultivars. However, this differential egg distribution should be taken into account to select insecticides used against codling moth egg. Some chemicals currently used to control codling moth eggs, such as some mineral oils, have been shown to lead to a different degree of control when applied on different oviposition substrates (fruit and leaves).

Egg distribution is random respect to individual fruits, but not respect to fruit clusters of different fruit density. Consequently, from the point of view of the oviposition site selection behavior of gravid females, no thinning practices seem to be useful for the control of the codling moth. Moreover, adult females do not discriminate among clusters with non-infested and codling moth infested fruit as oviposition site. However, under field conditions there is not egg-laying on ripe 'Bartlett' pears. Ethylene, a volatile produced and released by 'Bartlett' pears when they reach maturity, is likely to play a role. Ethylene, in the concentrations it is found in the intercellular spaces of ripe 'Bartlett' pears, affects adversely codling moth mating and it tends to decrease egg laying.

Selection of an oviposition site is affected by several factors. Traditionally, anatomical characteristics of oviposition substrates (pubescence) and release of volatile attractants and oviposition stimulants have been considered the key factors on the selection of an oviposition site. Results have shown that α -farnesene content in host fruits is correlated to preference as oviposition substrate on fruit surface. Preference tests for leaf surfaces showed the important role of anatomical characteristics of plant surfaces in oviposition site selection. Pubescent leaf surfaces (e.g., underside of 'Red Delicious' apple leaves) are avoided as oviposition sites. Smooth leaves of non-host species are also less preferred. This is related to chemical factors, such as lower content of oviposition stimulants (α -farnesene). However, our experiments have shown that other factors are equally crucial in the selection of oviposition sites by the adult codling moth females. Such factors include the position of the leaf surface, the orientation of the leaf surface relative to a fruit and to a light source, and the distance to a fruit.

CHAPTER 5**CODLING MOTH (LEPIDOPTERA: TORTRICIDAE)
EGG DEVELOPMENT AND MORTALITY
AND LARVAL BEHAVIOR AND MORTALITY
IN APPLE AND PEAR**

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5.1 Introduction

The codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), is a widely distributed pest of cultivated pome fruits (apple, pear, and quince), walnuts, and some stone fruits (Shel'Deshova 1967, Riedl 1983). Differences in susceptibility to codling moth damage have been observed among apple (Felt 1910, Newcomer & Yothers 1924, Cutright & Morrison 1935, Withead 1944) and pear (Westigard et al. 1976) cultivars. Differences in codling moth infestation levels among host species and cultivars may be a manifestation of host-specific behavior of the moth and larval stages or they may arise because differential mortality of immature stages on different hosts. Considerable progress has been made in the development of apple cultivars with resistance to diseases, especially apple scab, *Venturia inaequalis* (Cooke) Wint (Crosby et al. 1997), and some arthropod pests, such as woolly apple aphid, *Eriosoma lanigerum* (Hausman) (Homoptera: Aphididae), and the European red mite, *Panonychus ulmi* (Koch) (Acari: Tetranychidae) (White & Bush 1998). However, little is known about the basis of host resistance to the codling moth.

Among the codling moth life stages, the adult is the primary stage involved in host selection. In addition, the moth stage also selects the oviposition site. The codling moth deposits eggs on the upper and lower sides of leaves near fruit, on the fruit itself and also on woody tissue. The proportions of eggs laid on different plant parts vary with the host (Wood 1965, Westigard et al. 1976, Chapter 4). Characteristics of oviposition surfaces on different host cultivars could affect the development and survivorship of eggs and, consequently, lead to differences in susceptibility among host species and cultivars. Previous studies have focused on the effect of environmental variables (Lekic 1950, Wyniger 1956, Hagley 1972b) and natural enemies (MacLellan 1962, Ferro et al. 1975, Westigard et al. 1976) on codling moth egg mortality. However, no information is available on egg mortality on different hosts and plant surfaces which might explain, at least in part, why codling moth is more successful on some hosts but not on others. Hagley et al. (1980) suggested that the characteristics of the wax scales on some host surfaces, such as the upper side of pear leaves, could negatively affect egg laying. Characteristics of the wax scales can also affect the rate of air exchange between the leaf surface and the base of the codling moth

egg and, consequently, respiration of the egg stage. Presence of specific phyto-chemicals in the epidermis of fruit and leaves may affect development and survivorship of the egg stage.

After eggs hatch freshly emerged neonate larvae must locate and penetrate a fruit before they desiccate, starve, succumb to adverse weather conditions, or are consumed by predators (Ferro et al. 1975, Jackson 1982). The physiological state of neonate larvae as well as their searching efficiency speed influence their survival (Jackson 1976, Jackson 1982). Larvae may travel considerable distances before they reach a fruit (Steiner 1939). Hall (1934) suggested that neonates locate a fruit by random movement. However, a directed search would be more efficient and increase the likelihood for survival of the young larvae. Several authors have studied the orientation of neonate larvae to the food source (McIndoo 1928, 1929; Geier 1963, Landolt et al. 1998) but there is not agreement whether the search is random or directed. Neonate behavior, like adult behavior, may be affected by several cues including visual (McIndoo 1929), chemical/olfactory (McIndoo 1928, Sutherland 1972, Sutherland et al. 1974) and tactile (McIndoo 1928) stimuli from host fruits. Sutherland (1972), Sutherland & Hutchins (1973), and Russ (1976) showed that some volatile compounds (i.e., α -farnesene and its stereoisomers) act as larval attractants to the fruit, but the sphere of influence was only a few millimeters. It was suggested that codling moth larvae are attracted to apples due to orthokinesis (Landolt et al. 1998) and klinotaxis or tropotaxis (Bradley & Suckling 1995) in response to α -farnesene. Although these studies demonstrated that certain apple odors such as α -farnesene isomers aid codling moth larvae searching for fruit, the importance of visual cues is not clear. Moreover, there are not studies where the combined effect of visual and olfactory cues on larval behavior have been investigated.

Once a neonate larva arrives on the fruit surface, it must penetrate the epidermis to reach the inside of the fruit. Likelihood of survival is greatest for newly hatched larvae that quickly enter the fruit. Young larvae may engage in some exploratory feeding before penetration (Geier 1963, Jackson 1982). Presence of phyto-chemicals in the epidermis may signal to the larva to accept the fruit and proceed with entering it. Success of a young larva to enter the fruit may depend on the location where it attempts to enter (calyx end, side of

fruit) and on physical or chemical factors such as fruit firmness, pubescence of fruit, characteristics of the wax layer, and presence of noxious phyto-chemicals in the epidermis and sub-epidermal layers. Fragrance, skin firmness and degree of sweetness (subacidness) of the apple have been suggested as possible factors which determine susceptibility to codling moth attack (Cutright & Morrison 1935, Witehead 1940). Westigard et al. (1976) suggested that the presence of stone cells in some pears such as 'Anjou', 'Bosc', 'Comice' and 'Bartlett' provide a physical barrier to larval entry. Once inside the fruit the larva is essentially protected from predation and from the effects of adverse weather. However, rain has been indicated as causing drowning of first instar larvae inside the fruit (Hagley 1972b).

In many respects, pears represent a larval medium quite different from apples. Inside the fruit, larval mortality may occur because of the chemical and physical characteristics of the food source. Resistant factors in host fruits can limit larval establishment (antixenosis) and can also delay or prevent larval development (antibiosis).

The main objectives of this study were to determine the host-specific mortality of immature stages of the codling moth in apple and pear cultivars and to investigate the potential role of different factors conferring resistance to codling moth attack. Egg development and searching behavior of neonate larvae on different host cultivars were also investigated. A better understanding of the interaction of immature codling moth stages with different host cultivars will provide a rational basis for improved management of this pest and help growers adopt cultivar-specific control programs.

5.2 Materials and Methods

Codling moth population. A population of *C. pomonella* originally collected in 1991 in an apple block at the Mid-Columbia Agricultural Research and Extension Center (MCAREC), Hood River, OR, was maintained on thinning apples in an insectary at 24 ± 2.5 °C, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h as described by Riedl & Loher

(1980). This laboratory population supplied adults, eggs and larvae used in the experiments.

Research sites and plant material. Field experiments were conducted in an apple and in a mixed pear-apple-maple block located at the MCAREC. The apple block was 0.5 ha in size with a tree and row spacing of 6.1 m and was 31 years old. It consisted of alternating rows of 'Red Delicious' and 'Newtown' apple trees. The pear block was a 0.2-acre mixed planting of 35 year-old 'Anjou' and 'Bartlett' with a tree and row spacing of 8 m. Six-year-old 'Newtown' apple and three-year-old maple, *Acer platanifolia* cv. Emerald Queen, trees were interplanted in the tree row in 1998. Except for disease sprays and a normal fertilizer program trees in both blocks received no insecticide or acaricide treatments. Both blocks provided plant material for experiments. In addition to field experiments, behavioral tests with neonate larvae were carried out in a greenhouse under natural light under controlled temperature and relative humidity (27 ± 2.0 °C, $70 \pm 5\%$ RH). Larval experiments were conducted between 10 a.m. and 5 p.m. Preliminary experiments had shown that most eggs (80%) hatched during this period (S.M., unpublished data). Plant material for behavior experiments was collected about 10 minutes before use.

Egg development and mortality on different cultivars and oviposition surfaces. Egg mortality in the field was checked on fruit and foliage of 'Red Delicious' apple as well as 'Anjou' and Bartlett' pear. Fruit clusters of each cultivar were enclosed in cylindrical sleeve cages (30 cm long x 15 cm diameter). Five pairs of 1-day-old mated moths were released into a cage during one night. Before being left in the cage, the adults were kept in plastic vials for 24 h in the insectary and supplied with a 5% sucrose solution. During this period of time they mated. Before cages were installed, leaves and fruit were checked for eggs laid by feral moths. These eggs were removed. Water and 5% sucrose solution dispensed from cotton wicks were available to caged moths during the experiment. Moths were removed after one night and eggs laid on each surface counted and marked. Sleeve cages were left in place to exclude parasitoids and predators and additional egg laying by feral moths. The cohort of eggs on different plant parts was followed through embryonic development until hatch to estimate duration of development and mortality. Degree-day

(DD) requirements for egg development on different host cultivars and oviposition surfaces were compared. The experiment was conducted at three different times during the season (May 26 to June 8, July 24 to July 30, and August 15 to August 23) since characteristics of leaf and fruit surfaces change through the year. Each experiment consisted of three replications of about 30 eggs each for each cultivar, oviposition surface, and time period.

Larval orientation. Larval orientation experiments were conducted in small open wooden arenas (30 cm x 30 cm) whose surface was covered with wax paper. In these experiments the role of visual and olfactory stimuli from fruit in larval orientation were studied. The first experiment consisted of the simultaneous presentation of plant material (leaf and fruit) from different host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear). The different choices of fruit and leaves were placed randomly in a circle at the same distant to each other. Larvae less than 1 hour-old were released in the center of the arena. One larva was released at any one time. The number of larvae arriving and accepting a choice was recorded. A choice was considered 'accepted' when larvae remained on a leaf or fruit for at least 1 min. Two different experiments were conducted. Choices in the first experiment consisted of natural leaf (no stimuli from fruit), artificial fruit (only visual stimuli from fruit), and natural non-infested and codling moth infested fruit (visual and olfactory stimuli from fruit). Neonates were released at three different distances (3, 6, and 10 cm) from leaf and fruits. Tests were conducted with fruit measuring 18, 30, and 65 mm in diameter. Thirty larvae were released for each combination of host cultivar, fruit size, and distance. In the second choice test, the role of the shape and the color of the fruit in larval orientation was investigated. Similar procedures than in the previous experiment were followed, but in this case neonate larvae were exposed to artificial fruit (30 mm diameter) of different shape (apple and pear) and color (yellow, green, and red).

Larval travel speed. Larval travel speed was measured as the average linear speed of neonates to travel to a food source (fruit). Experiments were conducted with 20 cm long plastic rods (3 mm diameter) which were held horizontally at 20 cm from the surface. The plastic rod was attached on one end to a wooden post with an alligator clip hanging. Fruit was attached to the other end. Less than 1 hour-old neonate larvae were released

individually at the end of the rod where it was attached to the wooden post. The time needed for each neonate to travel to the fruit on the other end of the rod was recorded. Three different experiments were conducted. In the first experiment, the effect of visual and olfactory stimuli of host fruits ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear) on larval speed was studied. Natural non-infested, natural infested, or artificial fruit of different host cultivars of different stages of development (18, 30, and 65 mm of diameter) were attached to the free end of the plastic twig. Twenty larvae were used for each combination of cultivar, fruit treatment, and fruit size. Neonate larvae were released at 3 cm from the fruit. In the second experiment, the effect of the number of fruit (1, 2, and 3) and distance to a 'Red Delicious' fruit (3, 6, and 10 cm) on larval travel speed was studied. Twenty larvae were released for each combination of fruit density and distance to fruit. A third experiment was conducted to study the effect of branch surface characteristics of different cultivars on larval movement. A similar procedure was used but larval speed was measured on natural woody tissue (about 3 mm diameter) of 'Red Delicious', 'Anjou' and 'Bartlett' cultivars. A artificial fruit (30 mm diameter) was used as a visual stimulus at the free end of the branch. Lateral branches of twigs were removed since only linear speed was studied. Twenty larvae were released on branches of each cultivar.

Cultivar susceptibility to larval attack and larval entry sites. In 1998 and 1999 field experiments were conducted to measure the relative susceptibility of one apple cultivar, 'Red Delicious' apple, and 'Bartlett' and 'Anjou' pears, to first-instar codling moth entry. In 1998, individual codling moth eggs in the black head stage of development were cut from wax paper egg sheets and placed on fruit in the field. Small pieces (0.5 x 0.5 cm) of wax paper with one egg were attached to the fruit next to the petiole area with Elmer's Glue-All glue (Elmer's Products, Inabout Columbus, OH). A single egg was attached to each fruit. Individual fruits were covered with organdy bags (10 cm diameter x 15 cm long) to exclude other pests and to protect eggs from predation or attack by parasitoids. One hundred fruits of each cultivar were used each time. The test was repeated six times during the season from May to September. Fruits were checked periodically, and records were kept of the number of hatched eggs, successful entries, number of shallow surface feeding sites (stings), location of entries, and larval survivorship inside the fruit. In addition, 400 fruits of each

host cultivar were randomly checked to determine infestation levels in the field at three times during the season: beginning of the season (first week of June), end of egg hatching of the first generation (fourth week of July), and second generation (first week of September).

In 1999, additional field experiments were conducted with 'Anjou' and 'Bartlett' pears to determine how successful neonate larvae were entering fruit through the calyx area or through the side of the fruit. On one group of pears, the calyx area was covered with paraffin wax (PAROWAX™, Service Assets Corp., Newport Beach, CA) to force larvae to attempt penetration through the side of the fruit. On another group of pears, a #1 gelatine capsule (Eli Lilly & Company, Indianapolis, IN) with a single codling moth egg was placed over the calyx area to force neonate larvae to attempt penetration through the calyx area. The gelatine capsule was attached with paraffin wax to the fruit. A third group of pears with calyx area open served as control. Thirty fruit were used per treatment and the experiment was repeated three times during the season (from early June to late August). Fruits were checked after 2 weeks, and records were kept of the number of hatched eggs, and successful entries.

Additional experiments were conducted in 1999 to observe first-instar larval behavior on the surface of host fruits. The experiments were conducted in the greenhouse (27 ± 2.0 °C, $70 \pm 5\%$ RH). Individual neonate larvae were released on the fruit surface of host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pears). Records were kept on larval movement, sites of penetration attempts, location of entries, and approximate time required for entering through the side of the fruit or the calyx area. The experiments were conducted three different times during the season (May 29-31, July 1-3, and August 18-20).

Effect of starvation time on first-instar larval penetration. First-instar larvae were kept without water or food (in 5 cm diameter x 8 cm high plastic vials at 24 ± 2.5 °C, $60 \pm 5\%$ RH) for 0, 8, and 16 hours. Larvae were released individually on the surface of host fruits (60 mm diameter), near the petiole area, to study the effect of starvation time on larval behavior and mortality on different apple ('Red Delicious') and pear ('Anjou' and 'Bartlett') cultivars. The experiment was conducted in the greenhouse (27 ± 2.0 °C, $70 \pm 5\%$ RH) in August 6-12. Twenty larvae were used for each cultivar and starvation time combination.

Records were kept of the penetration success, locations of penetration attempts and actual entries, and approximate time to enter the fruit through the side of the fruit or the calyx area.

Determination of fruit characteristics. Skin and flesh firmness from a random sample of 10 fruits of each host cultivar ('Red Delicious', 'Anjou', and 'Bartlett') at each stage of development were recorded the same day that experiments began to assess cultivar susceptibility to larval attack. Firmness was determined by a University of California (UC) pressure tester with a 1-mm of diameter plunger. Skin firmness was determined by inserting the plunger 1 mm deep in fruit with skin intact. Flesh firmness was determined by inserting the plunger 5 mm deep in fruit with skin peeled off. In addition, photographic pictures were taken of longitudinal sections of host fruits to have a record of their internal anatomy.

Complexity of fruit-bearing branches, measured as the mean distance between lateral ramifications (leaf insertion, lateral branch), was determined from a random sample of ten two- year-old branches of each host cultivar.

Measurement of environmental variables and calculation of degree-days.

Temperature and relative humidity inside sleeve cages and organandy bags, as well inside the greenhouse, were recorded with HOBO StowAway loggers (Onset Instruments, Proccasset, MA, U.S.A.). Hourly temperatures inside the sleeve cages were used to calculate the degree-day requirements for completion of egg development. The lower and upper temperature thresholds considered were 10 and 31 °C, respectively. A horizontal cutoff was considered for the maximum temperature of development.

Statistical Analysis. Logistic regression for binomial responses (SAS 1990) was used to compare egg mortality on different plant surfaces and larval mortality on different fruit cultivars and at different starvation times. Chi-square tests (SAS 1990) in which the observed distribution of larvae was compared with an expected even distribution of settled larvae across all fruit treatments were used to compare the role of stimuli from different plant material on larval orientation. The same tests were used to compare preference site for penetration in different host cultivars. Percentages of larval mortality were transformed to arc sin and analyzed with two-sample *t*-test. Analysis of variance (ANOVA) was used to

compare egg development rates (DD) in different oviposition surfaces and host cultivars and to compare larval speed during fruit searching. ANOVA and two sample *t*-tests were carried out with the Number Cruncher Statistical System (Hintze 1997). Analyses of developmental rates were supported by survival analysis tests (Proportional Hazard Regression, PHREG) (SAS 1997). Untransformed means are reported in tables, figures, and text.

5.3 Results

Egg development and mortality on different cultivars and oviposition surfaces.

Natural mortality of the egg stage averaged 9% (Table 5.1). No differences on natural egg mortality were observed among host cultivars ($\chi^2 = 0.425$; $df = 2$; $P = 0.809$), oviposition surfaces (lower and upper part of the leaf, and fruit; $\chi^2 = 1.379$; $df = 2$; $P = 0.502$), and time of season ($\chi^2 = 1.301$; $df = 2$; $P = 0.522$). None of the two-factor interactions was significant ($\chi^2 < 0.90$; $df = 4$; $P > 0.90$).

Average duration of egg development ranged from 84.2 to 90.8 DD (Table 5.1).

There were statistically significant differences on the duration of the development of the egg stage among different host cultivars ($F = 6.338$; $df = 2$, 2266; $P = 0.002$) and oviposition surfaces ($F = 4.512$; $df = 2$, 2266; $P = 0.011$). Duration of the egg stage was also significantly different at the three times of the season ($F = 15.179$; $df = 2$, 2266; $P < 0.001$). The two-factor interaction 'cultivar x time' was significant ($F = 4.88$; $df = 4$, 2266; $P < 0.001$). Two-factor interactions 'surface x time' and 'surface x variety' were not significant. Egg development tended to be slower on the 'Anjou' cultivar, on fruit, and at the end of the season.

Table 5.1: Codling moth field egg mortality and heat requirements for completion of the egg stage on different host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear) and oviposition substrates at three periods of the 1998 season. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$).

Time of year	Cultivar	Plant surface	Egg mortality		Egg development (degree-days)	
			n	%	n	Mean (SEM)
5/26/98	'Red Delicious'	Fruit	101	7.9	93	88.7 (0.94) bc
		Underside of leaf	103	8.7	94	86.4 (0.77) bc
		Upper side of leaf	98	6.1	92	90.3 (0.84) a
	'Anjou'	Fruit	94	6.4	88	90.0 (0.79) a
		Underside of leaf	96	9.4	87	88.9 (0.69) abc
		Upper side of leaf	93	8.6	85	84.7 (0.91) ef
	'Bartlett'	Fruit	98	7.1	91	86.1 (0.93) ef
		Underside of leaf	87	8.0	80	88.7 (0.81) abcd
		Upper side of leaf	107	7.5	99	86.9 (0.65) cde
7/24/98	'Red Delicious'	Fruit	92	8.7	84	87.3 (1.07) bcde
		Underside of leaf	92	11.9	81	89.9 (0.87) ab
		Upper side of leaf	93	8.6	85	84.2 (0.91) f
	'Anjou'	Fruit	93	8.6	85	87.6 (1.01) bcde
		Underside of leaf	97	11.3	86	91.2 (1.00) a
		Upper side of leaf	91	9.9	82	89.9 (0.96) ab
	'Bartlett'	Fruit	88	8.0	81	90.3 (1.11) a
		Underside of leaf	102	9.8	92	85.2 (0.80) ef
		Upperside of leaf	91	8.8	83	86.6 (0.74) def
8/15/98	'Red Delicious'	Fruit	92	9.8	83	90.0 (0.74) ab
		Underside of leaf	93	11.8	82	90.2 (0.79) a
		Upper side of leaf	94	9.6	85	89.4 (0.67) abc
	'Anjou'	Fruit	84	9.5	76	89.6 (0.54) ab
		Underside of leaf	94	11.7	83	90.8 (0.69) a
		Upper side of leaf	88	10.2	79	89.9 (0.61) ab
	'Bartlett'	Fruit	83	9.6	75	90.0 (0.70) ab
		Underside of leaf	92	9.8	83	89.1 (0.49) abcd
		Upper side of leaf	88	10.2	79	90.1 (0.61) a

Larval orientation tests. Typically a neonate larva, after wandering in the arena for 20-30 seconds, started moving along in a particular direction until it stopped and raised the anterior half of its body into the air to test the surrounding atmosphere. Sometimes larvae continued in the same direction, but more often they made adjustments and changed course.

Neonate larvae showed the same behavior pattern when exposed to plant material of different host cultivars ($\chi^2 < 2.20$; $df = 6$; $P > 0.90$). Neonates avoided the leaf choice and moved towards the fruit choices, but they did not discriminate among natural non-infested, natural codling moth infested, and artificial fruit. The non-preference showed for different fruit treatments is the reason why statistical analyses showed that newly hatched larvae released in the arena moved in an apparent random manner ($P > 0.05$; Table 5.2).

Significant departure from randomness was only observed when larvae were exposed to 18 mm of diameter 'Anjou' pear at 3 cm ($\chi^2 = 9.507$; $df = 3$; $P = 0.023$), to 30 mm of diameter 'Bartlett' pear at 6 cm ($\chi^2 = 8.188$; $df = 3$; $P = 0.042$), and to 65 mm of diameter 'Red Delicious' apple and 'Bartlett' pear at 3 cm of distance ($\chi^2 = 9.477$; $df = 3$; $P = 0.024$; and ($\chi^2 = 9.261$; $df = 3$; $P = 0.026$, respectively)

Neither color nor shape of fruit were critical factors for larval orientation at any distance ($\chi^2 < 0.556$; $df = 5$; $P > 0.99$) (Table 5.3).

Table 5.2: Percentage of codling moth first instar larvae responding to fruit and leaf choices of different host cultivars ('Red Delicious' apple, and 'Anjou' and 'Bartlett' pear) in preference tests (random searching if $P > 0.05$).

Fruit diameter (mm)	Cultivar		Distance of release point to fruit or leaf (cm)		
			3	6	10
18	'Red Delicious'	% on natural non-infested fruit	23.3	30.0	30.0
		% on natural infested fruit	43.3	26.7	26.7
		% on artificial fruit	30.0	33.3	20.0
		% on leaf	3.3	10.0	23.3
		Chi-square test for randomness (df = 3)	$\chi^2 = 6.888; P = 0.076$	$\chi^2 = 3.052; P = 0.384$	$\chi^2 = 0.384; P = 0.942$
	'Anjou'	% on natural non-infested fruit	26.7	30.0	26.7
		% on natural infested fruit	43.3	26.7	30.0
		% on artificial fruit	30.0	26.7	30.0
		% on leaf	0.0	16.7	13.3
		Chi-square test for randomness (df = 3)	$\chi^2 = 9.507; P = 0.023$	$\chi^2 = 1.008; P = 0.799$	$\chi^2 = 1.708; P = 0.636$
	'Bartlett'	% on natural non-infested fruit	30.0	30.0	26.7
		% on natural infested fruit	40.0	33.3	30.0
		% on artificial fruit	26.7	26.7	20.0
		% on leaf	3.3	10.0	23.3
		Chi-square test for randomness (df = 3)	$\chi^2 = 6.561; P = 0.087$	$\chi^2 = 2.812; P = 0.422$	$\chi^2 = 0.288; P = 0.966$
30	'Red Delicious'	% on natural non-infested fruit	23.3	26.7	30.0
		% on natural infested fruit	40.0	30.0	30.0
		% on artificial fruit	26.7	36.7	26.7
		% on leaf	10.0	6.6	13.3
		Chi-square test for randomness (df = 3)	$\chi^2 = 3.138; P = 0.371$	$\chi^2 = 4.614; P = 0.202$	$\chi^2 = 1.708; P = 0.636$
	'Anjou'	% on natural non-infested fruit	26.7	33.3	26.7
		% on natural infested fruit	43.3	30.0	26.7
		% on artificial fruit	23.3	30.0	26.6
		% on leaf	6.7	6.7	20.0
		Chi-square test for randomness (df = 3)	$\chi^2 = 4.857; P = 0.183$	$\chi^2 = 4.438; P = 0.218$	$\chi^2 = 0.418; P = 0.936$
	'Bartlett'	% on natural non-infested fruit	23.3	40.0	26.7
		% on natural infested fruit	53.3	30.0	23.3
		% on artificial fruit	20.0	26.7	30.0
		% on leaf	3.3	3.3	20.0
		Chi-square test for randomness (df = 3)	$\chi^2 = 8.188; P = 0.042$	$\chi^2 = 6.888; P = 0.076$	$\chi^2 = 0.668; P = 0.880$
65	'Red Delicious'	% on natural non-infested fruit	33.3	30.0	30.0
		% on natural infested fruit	30.0	30.0	30.0
		% on artificial fruit	36.7	36.7	33.3
		% on leaf	0.0	3.3	6.7
		Chi-square test for randomness (df = 3)	$\chi^2 = 9.477; P = 0.024$	$\chi^2 = 6.642; P = 0.084$	$\chi^2 = 4.438; P = 0.218$
	'Anjou'	% on natural non-infested fruit	30.0	30.0	36.7
		% on natural infested fruit	30.0	26.7	30.0
		% on artificial fruit	36.7	40.0	30.0
		% on leaf	3.3	3.3	3.3
		Chi-square test for randomness (df = 3)	$\chi^2 = 6.642; P = 0.084$	$\chi^2 = 7.010; P = 0.072$	$\chi^2 = 6.642; P = 0.084$
	'Bartlett'	% on natural non-infested fruit	33.4	30.0	33.3
		% on natural infested fruit	33.3	33.3	26.7
		% on artificial fruit	33.3	33.3	33.3
		% on leaf	0.0	3.4	6.7
		Chi-square test for randomness (df = 3)	$\chi^2 = 9.281; P = 0.028$	$\chi^2 = 6.446; P = 0.082$	$\chi^2 = 4.668; P = 0.199$

Table 5.3: Percentage of codling moth first instar larvae responding to artificial fruits of different color and shape (random searching if $P > 0.05$).

	Distance of release point to fruit (cm)		
	3	6	10
%on green apple	20.0	23.3	20.0
%on red apple	13.2	16.7	16.7
%on yellow apple	16.7	16.7	13.2
%on green pear	16.7	16.7	16.7
%on red pear	16.7	13.3	16.7
%on yellow pear	16.7	13.3	16.7
Chi-square test for randomness (df = 5)	$\chi^2 = 0.212, P = 0.999$	$\chi^2 = 0.555, P = 0.990$	$\chi^2 = 0.212, P = 0.999$

Larval travel speed. Larval linear speed averaged 2.44 cm/min and ranged from 0.79 cm/min to 4.29 cm/min (Table 5.4). Average linear speed was not significantly different when larvae were exposed only to visual stimuli (artificial fruit) and to visual and olfactory stimuli from natural non-infested and codling moth infested fruit ($F = 0.09$; $df = 2, 513$; $P = 0.910$; Table 5.4). Larval speed was the same when neonates moved towards fruit of different hosts ($F = 0.49$; $df = 2, 513$; $P = 0.613$) and of different sizes ($F = 1.14$; $df = 2, 513$; $P = 0.319$; Table 5.4). Neither distance to the fruit ($F = 0.72$; $df = 2, 171$; $P = 0.486$) nor the number of fruit/cluster ($F = 1.05$; $df = 2, 171$; $P = 0.352$) affected larval linear speed to reach the fruit (Table 5.4).

Average larval linear speed was not affected by the surface characteristics of the branches of different host cultivars tested ($F = 0.11$; $df = 2, 51$; $P = 0.893$) (Table 5.4). Branches from pear cultivars have a slightly smoother surface than apple branches, but no differences in complexity of branches (lateral ramifications) were observed in different host cultivars ($F = 0.57$; $df = 2, 27$; $P = 0.570$).

Table 5.4: Mean linear speed of first instar codling moth larvae exposed to artificial and natural host fruits, at three stages of fruit development, at different distances from fruit, and on branches of different host cultivars. Means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Fruit size	No. of fruit	Distance to fruit	Surface	Fruit treatment	Cultivar	Larval speed: Mean (SEM) (cm/min)
18 mm	1	3 cm	Artificial twig	Artificial fruit	Red Delicious	2.64 (0.17) ns
					Anjou	2.47 (0.17)
					Bartlett	2.44 (0.16)
				Natural non-infested	Red Delicious	2.33 (0.13)
					Anjou	2.42 (0.17)
					Bartlett	2.36 (0.14)
				Natural infested	Red Delicious	2.35 (0.18)
					Anjou	2.25 (0.13)
					Bartlett	2.50 (0.16)
30 mm	1	3 cm	Artificial twig	Artificial fruit	Red Delicious	2.39 (0.14)
					Anjou	2.33 (0.09)
					Bartlett	2.33 (0.09)
				Natural non-infested	Red Delicious	2.37 (0.12)
					Anjou	2.49 (0.15)
					Bartlett	2.66 (0.20)
				Natural infested	Red Delicious	2.42 (0.14)
					Anjou	2.37 (0.11)
					Bartlett	2.58 (0.19)
60 mm	1	3 cm	Artificial twig	Artificial fruit	Red Delicious	2.48 (0.13)
					Anjou	2.44 (0.16)
					Bartlett	2.44 (0.16)
				Natural non-infested	Red Delicious	2.59 (0.09)
					Anjou	2.48 (0.14)
					Bartlett	2.56 (0.16)
				Natural infested	Red Delicious	2.57 (0.14)
					Anjou	2.55 (0.14)
					Bartlett	2.53 (0.14)
30 mm	1	3 cm	Artificial twig	Natural fruit	Red Delicious	2.49 (0.14)
	2					2.43 (0.12)
	3					2.31 (0.15)
30 mm	1	6 cm	Artificial twig	Natural fruit	Red Delicious	2.39 (0.12)
	2					2.44 (0.14)
	3					2.30 (0.10)
30 mm	1	10 cm	Artificial twig	Natural fruit	Red Delicious	2.69 (0.14)
	2					2.34 (0.18)
	3					2.49 (0.15)
30 mm	1	3 cm	Natural twig	Artificial fruit	Red Delicious	2.33 (0.16)
					Anjou	2.36 (0.14)
					Bartlett	2.43 (0.16)

Cultivar susceptibility to larval attack and larval entry sites. Significant differences in first instar larval mortality were observed over most of the season among different cultivars. Differential first instar mortality was primarily due to the differences in success of neonates to penetrate fruit on different host ($\chi^2 = 54.479$; $df = 2$; $P < 0.001$; Fig. 5.1). First instar larval mortality was significantly different through the season ($\chi^2 = 41.264$; $df = 5$; $P < 0.001$). Differences on penetration success between cultivars varied through the season ($\chi^2 = 51.674$; $df = 10$; $P < 0.001$, for the interaction term 'cultivar x time of season').

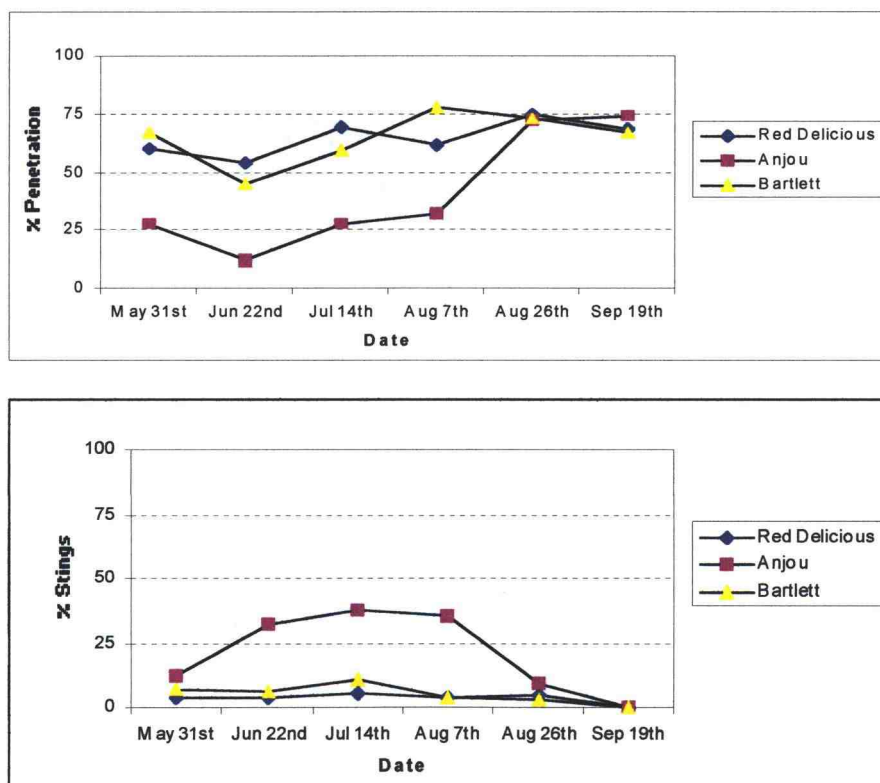


Fig. 5.1: Relative susceptibility of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars to entry by first instar codling moth larvae during the 1998 season.

Mortality was significantly higher on 'Anjou' (> 70%) than on 'Bartlett' pear and 'Red Delicious' apple (around 40%) (Fig. 5.1), except at the end of the season. When pears were close to maturity the percentage of first instar larvae successfully entering fruit was higher than at any time during the season and similar among the different cultivars (about 70%) (Fig. 5.1). The proportion of fruits with shallow feeding scars (stings) in different cultivars was inversely correlated with penetration success and suggested increased wandering and multiple attempts to enter less susceptible fruit (Fig. 5.1).

Infestation levels in different host cultivars (Fig. 5.2) showed a pattern of susceptibility similar to that observed in field experiments with larvae from the laboratory colony.

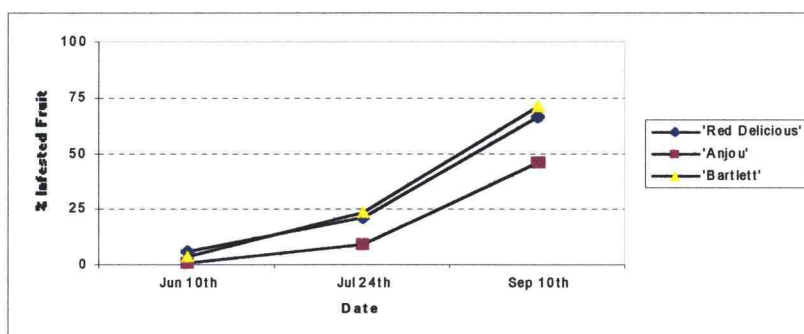
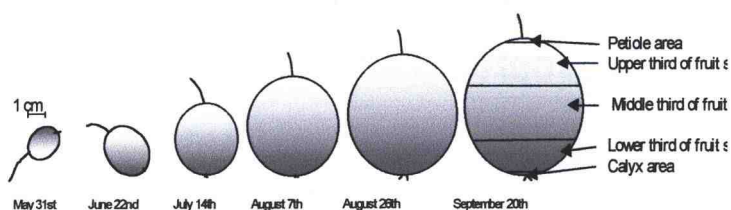
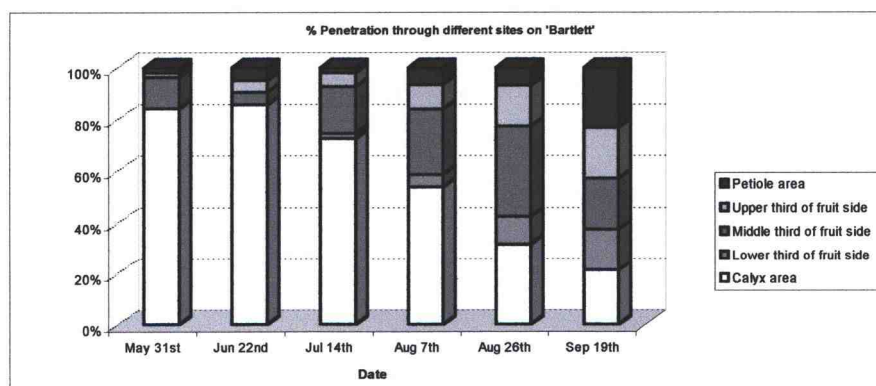
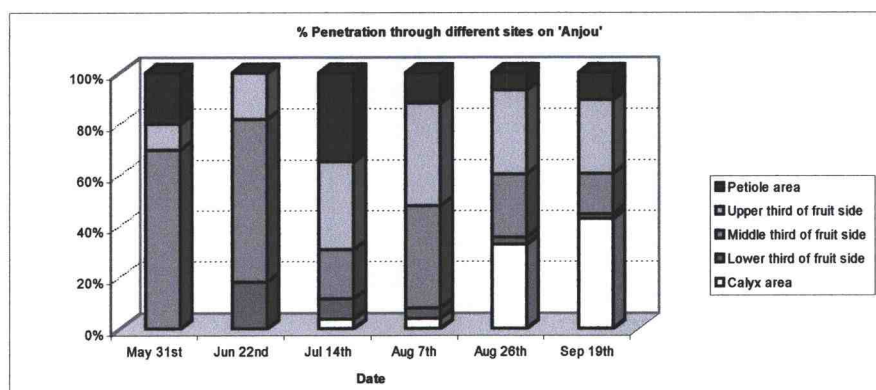
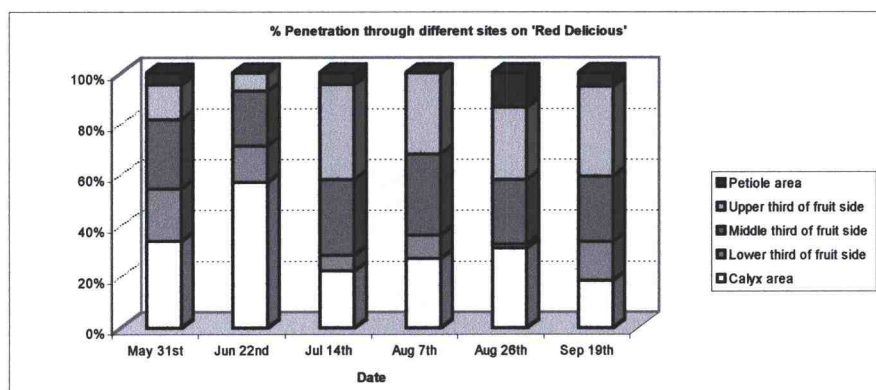


Fig. 5.2: Relative infestation by codling moth of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars in the field during the 1998 season.

There were significant differences in the penetration site in different host cultivars during most of the season ($\chi^2 > 30.0$; $df = 8$; $P < 0.001$) (Fig. 5.3). Differences were not significant when fruit approached maturity ($\chi^2 = 15.422$; $df = 8$; $P = 0.051$). In 'Anjou' pears, neonate larvae were unable to penetrate through the calyx area, except when fruit was almost mature, and larvae penetrated predominantly through the side of the fruit (Fig. 5.3). In 'Bartlett' pears, the calyx area (including the area just beneath the calyx lobes) was the preferred penetration site (70-80%) until late in the season. No preference for specific penetration sites was observed in 'Red Delicious' fruit (Fig. 5.3).



Fruit firmness might have some effect on penetration success. Skin and flesh firmness of 'Red Delicious' apple, and 'Anjou' and 'Bartlett' pear fruits are shown in Fig. 5.4. 'Red Delicious' apple had lower skin firmness than pears until mid-season and lower flesh firmness during the whole season. 'Bartlett' fruits had skin and flesh firmness as high as 'Anjou' pears, except at the end of the season.

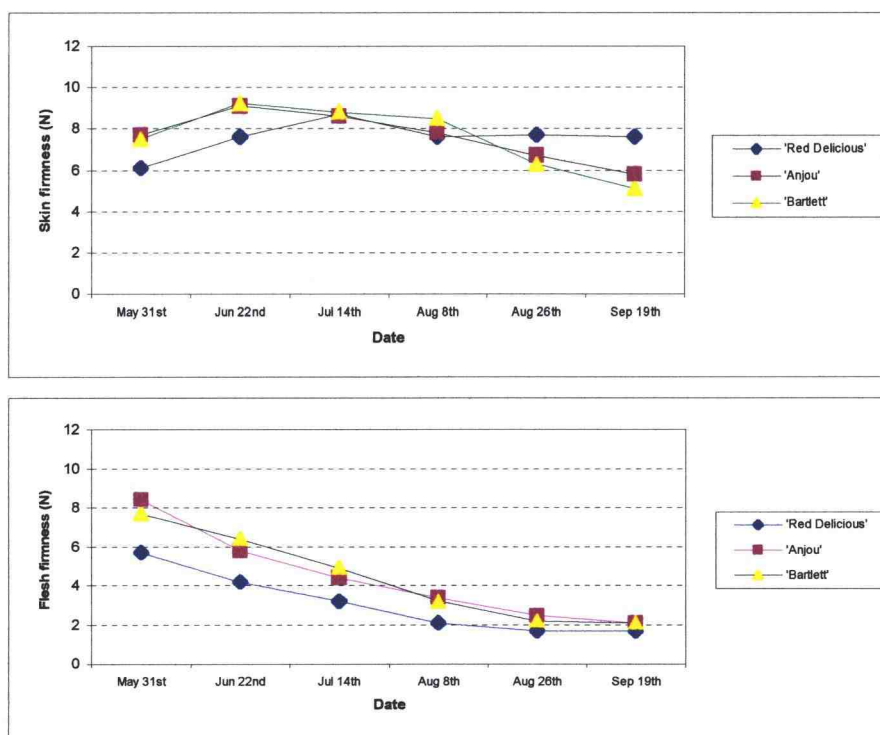


Fig. 5.4: Skin and flesh firmness on fruits of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars through the 1998 season.

The stage of development of the fruit affected first-instar larval behavior on the surface of host fruits (Fig. 5.5). Early in the season and in all host fruits (15-20 mm diameter) most neonate larvae went directly to the calyx area and attempted to penetrate there. At the middle of the season (30-35 mm fruit diameter), neonate larvae wandered more on the side of the fruit surface, where they tried penetration. However, in pears, which

generally had a higher skin firmness (Fig. 5.4), larvae ended up attempting penetration mostly through the calyx area (66.7 % on 'Anjou' and 64.3 % on 'Bartlett') after the first attempt to penetrate through the side of the fruit (Fig. 5.5). Later in the season (65 mm fruit diameter), most neonate larvae tried to penetrate directly through the side of the fruit (Fig. 5.5).

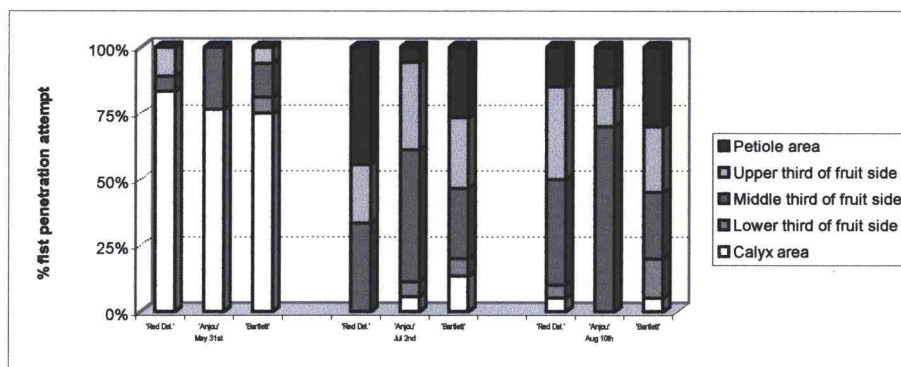


Fig. 5.5: Site of first penetration attempt of first instar codling moth larvae on one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars at different times of the 1999 season.

An additional experiment conducted in the field with 'Bartlett' and 'Anjou' pears showed that differences in first-instar larval mortality at the beginning of the season were due to the inability of neonate larvae to penetrate through the calyx area of 'Anjou' pears. Only 3.4 % of neonates that were forced to attempt penetration in the calyx area were able to enter the fruit (Fig. 5.6). Skin firmness on both cultivars was similar at that period as was the ability of neonate to penetrate through the side of the fruit (about 30%; Fig. 5.6). Differences on penetration success were probably related to anatomical characteristics. Pictures of longitudinal sections of fruits showing some aspects of the internal anatomy of 'Red Delicious' apple and 'Anjou', 'Bosc', and 'Bartlett' pears are presented in Fig. 5.7. Apple fruits do not have stone cells, but both summer and winter pear cultivars have a layer of stone cells. In 'Anjou' fruits, as well as in other winter pear cultivars such as 'Bosc', the

stone cell layer completely surrounds the interior of the fruit, including the calyx area. Small 'Bartlett' fruits also have a layer of stone cells. However, the calyx area of 'Bartlett' pears is free of stone cells (Fig. 5.7). This discontinuity, or 'channel', allows neonate larvae to penetrate to the fruit interior. The thickness of the layer of stone cells decreased as the season advanced.

At the middle of the season, in 'Anjou' pears neonate larvae were still quite unable to penetrate both through the calyx (13.3%) and through the side of the fruit surface (20.8%). In 'Bartlett' pears, larvae that were forced to attempt penetration through the calyx area were still the most successful entering the fruit (73.3%). Even though the skin and flesh firmness were slightly higher on 'Bartlett' than on 'Anjou' pears, penetration rates through the side of the fruit surface increased considerably in 'Bartlett' pears where larvae were forced to attempt penetration through the side of the fruit (60.0%).

The calyx area was not the preferred penetration site as fruit matured. At the end of the season, in 'Anjou' pears first-instar larvae were able to penetrate at a much higher rate through the site of the fruit (60.0%), but their ability to penetrate through the calyx was still low (20.7%). In 'Bartlett' pears, penetration success of neonate larvae was also higher through the side of the fruit surface (76.7%) than through the calyx area (51.9%; Fig. 5.6).

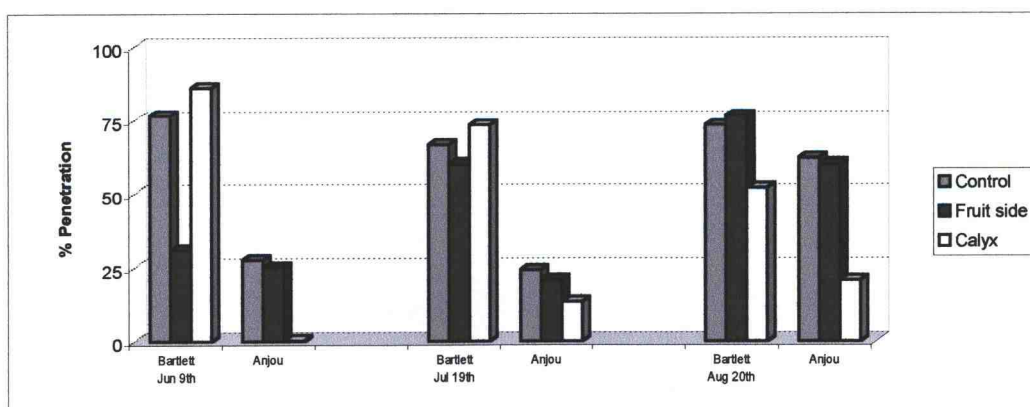


Fig. 5.6: Relative susceptibility of 'Anjou' and 'Bartlett' pears to entry by first instar codling moth larvae through different sites of the fruit at different times of the 1999 season.

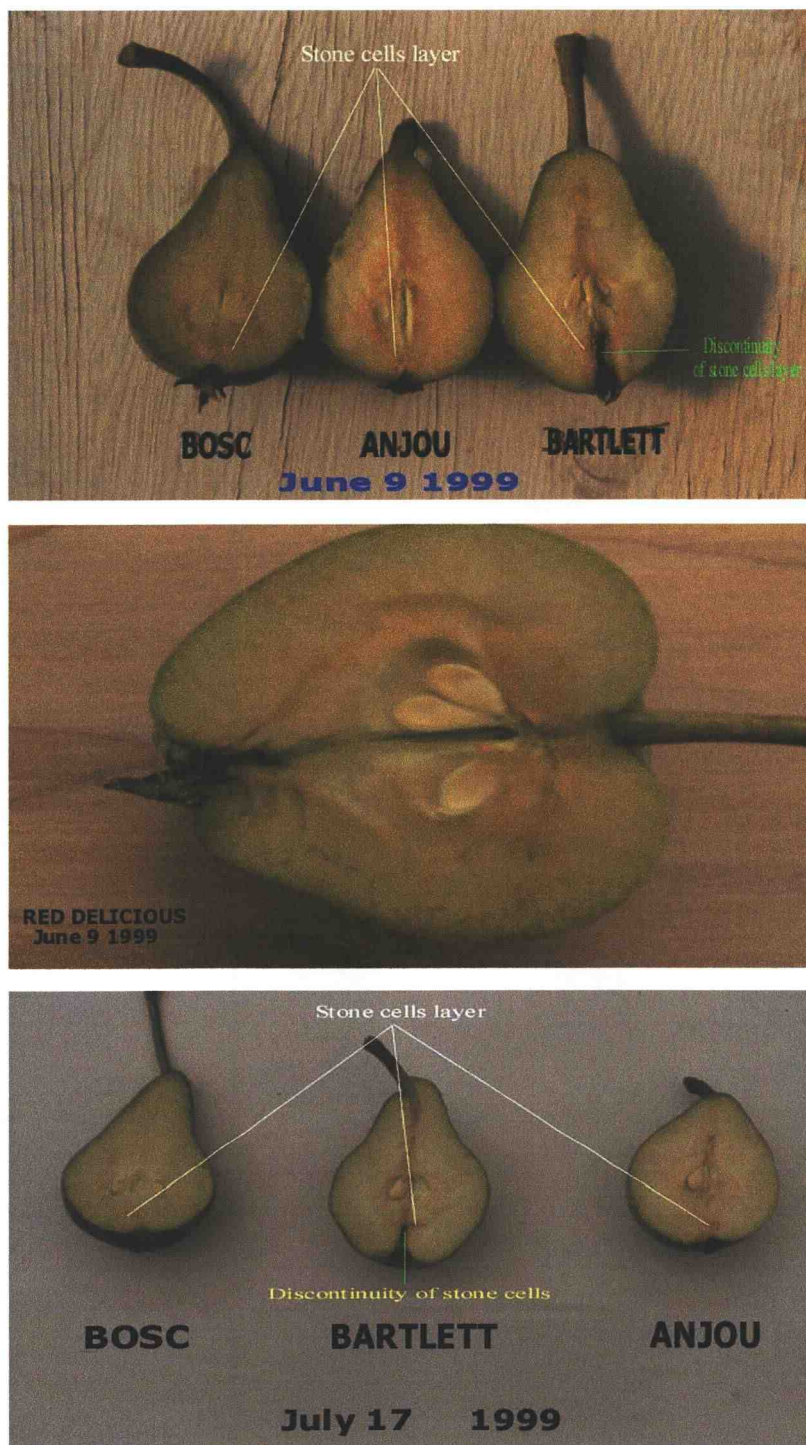


Fig. 5.7: Photographic pictures of longitudinal sections of host fruits ('Red Delicious' apple, and 'Anjou', 'Bosc', and 'Bartlett' pear) showing some aspects of their internal anatomy.

Mortality of larvae inside the fruit was around 10% for most of the season (Fig. 5.8) and was significantly lower during the whole season compared to first instar mortality before penetration ($t > 3.8$, $df = 4$, $P < 0.01$). There were no significant differences in terms of larval mortality among the different host cultivars after neonates had successfully entered the fruit ($\chi^2 = 3.789$; $df = 2$; $P = 0.150$; Fig. 5.8). However, larval mortality inside the fruit was higher (around 25%) early and late in the season than in the middle of the season (around 10%) in all host cultivars ($\chi^2 = 48.080$; $df = 4$; $P < 0.001$; Fig. 5.8). Larval mortality inside the fruit occurred during the first and second instars.

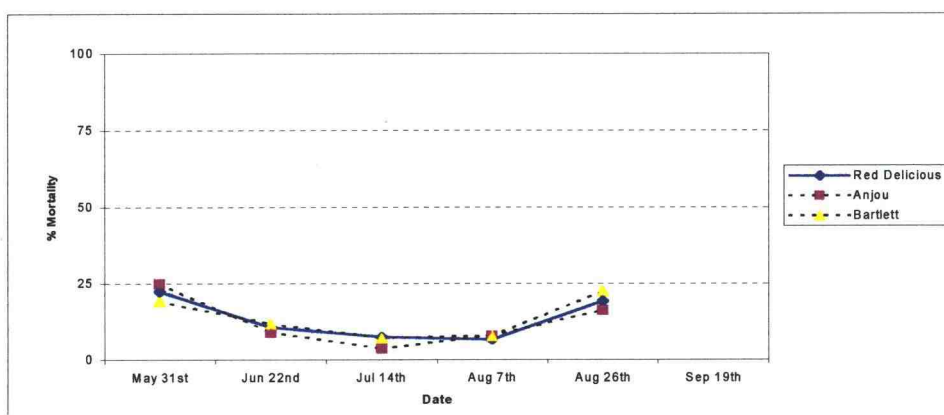


Fig. 5.8: Codling moth larval mortality inside the fruit of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars through the 1998 season.

Effect of starvation time on first-instar larval penetration. The time between egg hatching and fruit entrance attempt (starvation time) affected both first instar larval mortality ($\chi^2 = 15.112$; $df = 2$; $P = 0.0005$) and larval behavior on the fruit surface. Penetration success (Fig. 5.9) and time to reach the inside of the fruit were inversely related to starvation time in the three host cultivars tested. However, there was also a cultivar effect

($\chi^2 = 19.703$; $df = 2$; $P = 0.0001$). The success of newly hatched larvae to penetrate fruit (80.0% in 'Red Delicious', 70.0% in 'Bartlett', and 25.0% in 'Anjou') was similar to penetration success under field conditions at that time of the season (first third of August; Fig. 5.1). Larvae spent more time exploring the fruit surface and there was more variability in the site where neonates attempted to penetrate. Penetration success decreased when larvae were starved for 8 hours. The decrease in penetration success was especially noticeable in pears (35.0% in 'Bartlett', and 15.0% in 'Anjou'). When larvae were starved for 16 hours, most larvae ceased to explore the fruit surface and tried to penetrate where they were released (next the petiole area). Penetration success was very low (< 35.0%) in all host cultivars. With 'Red Delicious' apple a significant decrease in penetration success was observed compared to just 8 hours of starvation. No larvae survived after 24 h of starvation at 24 ± 2.5 °C, $60 \pm 5\%$ RH.

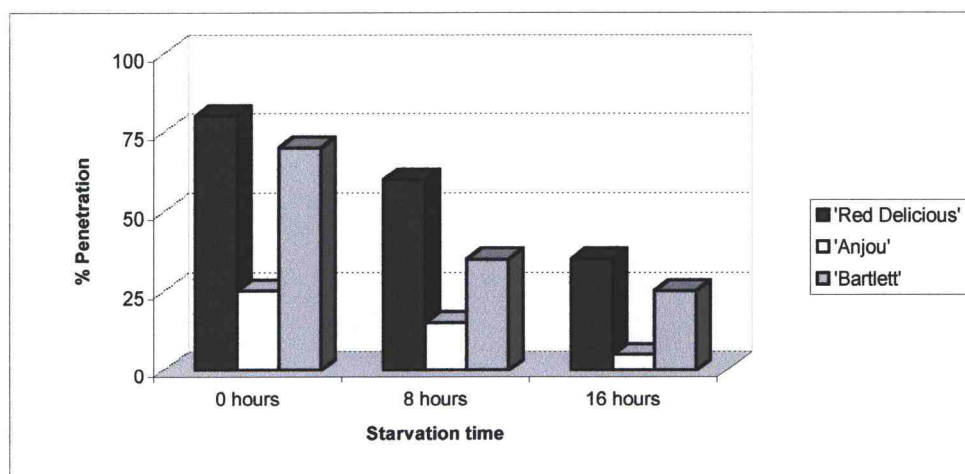


Fig. 5.9: Relative susceptibility of one apple ('Red Delicious') and two pear ('Anjou' and 'Bartlett') cultivars to entry by first instar codling moth larvae kept under starvation conditions during different periods of time.

5.4 Discussion

Natural egg mortality (around 9%) was similar to that observed by Wood (1965) who found 25% egg mortality with 14% attributed to parasitism on unsprayed 'Gravenstein' apple and 4.7-6.9% on 'Delicious' and 'Sturmer' apples. Similar results on unsprayed 'Bartlett' pear were reported by Westigard et al. (1976) who found 26% egg mortality with 10% attributed to parasitism. Higher rates of natural egg mortality (25%) were found by Ferro et al. (1975) in cage studies in an apple orchard where predation and parasitism were excluded. Results suggest that the natural oviposition substrate regardless of cultivar had no effect on egg mortality. Therefore, differential egg mortality among hosts does not contribute to observed differences in susceptibility to codling moth among apple and pear cultivars.

There were statistically significant differences of the average duration of the egg stage among host cultivars, oviposition substrates, and times of the season. However, these differences (not greater than 6 degree-days) are not important biologically and from a practical point of view. Less than one day with moderate or high temperatures is required to accumulate 6 degree-days, so these differences would not affect the decision making for the control of the pest on the egg stage. Duration of the egg stage (84.2 to 90.8 degree-days) was similar to that observed by Pitcairn et al. (1991) who reported requirements of 94 degree-days for completion of the egg development under constant temperatures. Martí et al. (1996) reported similar heat requirements (88-90 degree-days) for the development of the egg stage of a Spanish strain under semifield conditions.

In multiple-choice experiments, newly hatched larvae released in an arena moved in an apparent random manner. Larvae avoided the leaf choice but did not discriminate among artificial (just visual stimuli) and natural (both visual and olfactory stimuli) fruit. Neither color nor shape of fruit were critical factors for larval orientation. Several authors (McIndoo 1929, Hall 1934, Jackson 1976) reported that neonate larvae move randomly after hatching. Other workers thought that larval orientation is more directed (Geier 1963) and that olfaction is an important factor guiding first instar larvae to the fruit, especially at short

distances (Sutherland 1972). Sutherland & Hutchins (1972, 1973), Bradley & Suckling (1995) and Landolt et al. (1998) demonstrated that larval codling moth attraction may be related to some volatile components (i.e. α -farnesene) from apple fruits. The non-preference for natural over artificial fruit choices does not exclude the role of chemical stimuli in the orientation of neonate larvae at very short distances, but suggest that both the photosensitive behavior of neonate larvae and visual stimuli from fruit are more important than olfactory stimuli from fruit for larval orientation. Tactile stimuli did not seem to be critical for fruit acceptance since artificial fruits were equally accepted once contacted. Jackson (1982) suggested that the net result of the neonate larvae positive phototropism could be crucial to find the fruit, leading neonate from inner, darker, portions of branches to their apices where fruits are normally found. The same author reported no discrimination among natural non-infested and natural infested (attacked by just one larva) apple fruits by neonate larvae. Previous work testing the role of olfactory stimuli, basically α -farnesene (Bradley & Suckling 1995, Landolt et al. 1998), has not combined olfactory and visual cues from fruit. Therefore, the relative role of visual and olfactory stimuli from fruits could not been investigated. Moreover, experiments conducted in olfactometers with airflow (i.e. Landolt et al. 1998) may create artificial conditions. Wind conditions (speed, direction) change constantly in the field and the distribution of volatile concentrations and gradients within a host tree is unknown. Chemoanemotactic responses (chemical modulation of responses to wind) are nearly always involved in insect attraction to chemical volatiles, especially long range attraction to adults (Kennedy 1977), and there is a need to understand orientation to volatilized chemicals in moving air. However, it cannot be assumed that the behavior observed in those experiments is representative of what happens in nature. Russ (1976) observed that neonate larvae significantly preferred apple fruit extract as well as ripe apple fruit as compared to pear extract or ripe pear fruit in tests conducted in petri-dish arenas. He did not mention what cultivars he used in his tests and he assumed that α -farnesene content was higher in apples than in pears. Analyses on fresh fruit had shown that ripe pears (both 'Anjou' and 'Bartlett') have a higher content of α -farnesene than ripe 'Red Delicious' apples (Table 3.1; Chapter 3). So there are contradictory results about the role of this volatile on larval orientation. A better knowledge of the conditions (volatile concentrations, gradients) to which neonate larvae are exposed in host trees is needed. This

information could help to design better larval searching experiments in controlled conditions. In addition, there is always the possibility that chemical cues might have a more important role guiding neonate larvae of field populations.

Larval linear speed averaged 2.44 cm/min and ranged from 0.79 cm/min to 4.29 cm/min. Hall (1934) and Jackson (1982) reported similar larval speeds under comparable temperature conditions.

Average linear speed was not significantly different when larvae were exposed only to visual stimuli to both visual and chemical stimuli from natural non-infested and infested fruit. Bradley & Suckling (1995) and Landolt et al. (1998) suggested that larval orientation to apple odor could involve an increase in forward speed (orthokinesis). McIndoo (1928) hypothesized that olfactory stimuli from fruit affect larval behavior only within a few millimeters of the food source. A concentration gradient and threshold might be necessary to trigger orthokinetic responses in neonate larvae. These conditions likely did not occur in our experiments and probably are not present in nature except within a few millimeters of the fruit. Consequently, orthokinetic responses in first instar larvae may not affect average speed when measured at much longer distances. Data suggest that fruit odors have a negligible effect or their area of influence is too limited to affect average larval linear speed in the range of distances most eggs are laid from a fruit under field conditions (1 to 6 cm).

Larval average linear speed was not affected by the surface characteristics of the branches of different host cultivars. As pointed by Jackson (1982) and Subinprasert (1988), since neonate larvae show random movement, the number of ramifications on twigs containing fruit clusters could be directly related to the number of larvae reaching the fruit and time needed to arrive to the fruit and, consequently, to first instar larval mortality. Results and characteristics of branches of different host cultivars suggest that differences in codling moth infestation levels among host cultivars can not be attributed to differential larval behavior while searching for food.

Significant differences in first instar larval mortality were observed over most of the season among different cultivars. These differences in first instar mortality were due to the different success to penetrate fruit in different host cultivars. Penetration success had the

same seasonal pattern than those reported by Westigard et al. (1976). Penetration rates in pear were higher, except at the end of the season, compared with the results reported by Westigard et al. (1976). Penetration success in apple was lower compared with the results reported by Westigard et al. (1976). These authors conducted the experiment in the laboratory with collected fruit. Pear fruits dehydrate fast once collected from the tree. This could explain the lower penetration success observed by these authors. This faster dehydration of pear fruits is probably related to a thinner wax layer compared with apple fruits. In addition, they used 'Yellow Delicious' apple, that is more susceptible to codling moth attack than 'Red Delicious'.

Infestation levels observed in the field were positively correlated to fruit penetration success in experiments with larvae from the laboratory colony. These results suggest that differential susceptibility to fruit penetration by first instar larvae might be the basis for resistance to codling moth attack among host cultivars.

Preference for penetration site on 'Bartlett' pears was similar to that reported by Westigard et al. (1976) who observed 88% of larval entries through the calyx or base of sepals and only 10% through the side of the fruit during two consecutive seasons. Provancher (1874) and Huard (1916) observed that larvae entered the fruit mostly by the calyx area, but Mailloux & LeRoux (1960) reported that larvae mostly preferred the side of the fruit to penetrate apple fruits.

Results suggest that a combination of several factors, including unique physical (skin firmness), anatomical (stone cells) and likely chemical features of fruits, as well as the behavior of neonate larvae on fruit surface, could explain the differences in mortality among host cultivars through the season. Fruit firmness was lower in 'Red Delicious' than in both pear cultivars until mid-season. This could explain why neonate larvae were more successful penetrating through the side of the fruit in 'Red Delicious' apple than in any pear fruit. However, there was no strong correlation between skin firmness and first instar larval mortality in pear cultivars: penetration success was higher in 'Bartlett' than in 'Anjou' fruit, but skin and flesh firmness were as high in 'Bartlett' as in 'Anjou' pears, except at the end of the season. This suggests that other factors must be involved in fruit resistance to codling moth larvae entry in 'Anjou' pears. Neonate behavior on the fruit surface and anatomical

characteristics of the fruit can explain the observed differences in penetration site and success among pear cultivars.

The stage of development of the fruit affected first-instar larval behavior on the fruit surface. Early in the season neonate larvae mostly moved to the calyx area and attempted to penetrate there. This behavior could be related to (1) a positive phototaxis of neonate larvae (small fruit has the calyx area oriented upwards), (2) chemical attractants from the calyx area, and/or (3) a negative response to gravity. McIndoo (1929) did not find evidence that first instar larvae are influenced by gravity. Jackson (1982) observed that larval movement was generally upwards but he could not associate this behavior to a response to gravity. At the middle of the season neonate larvae wandered more on the fruit side surface. However, in pears larvae ended up attempting penetration mostly through the calyx area after the first attempts on the side of the fruit. Consequently, the ability of neonate codling moth larvae to penetrate through the calyx area of some pear cultivars ('Bartlett') but not others (winter varieties such as 'Anjou' and 'Bosc') could help explain the observed differences in larval mortality, especially during early and mid-season.

Differences on the ability to enter the fruit were probably related to anatomical characteristics. The key factor explaining differences on penetration success among pear cultivars could likely be the different anatomy of the layer of stone cells. Stone cells are sclerenchyma tissue cells with a greatly thickened cell wall that provide support and mechanical strength (Ville 1967). In 'Anjou' fruits, as well as in other winter pear cultivars such as 'Bosc', the layer completely surrounds the interior of the fruit, including the calyx area. 'Bartlett' fruits have a discontinuity of the layer of stone cells in the calyx area. This 'channel' allows neonate larvae to penetrate to the fruit interior. At mid-season, penetration success through the side of the fruit was also higher in 'Bartlett' than in 'Anjou' pears. Consequently, at this time of the season, differences on 1st instar larval mortality were related to the differential ability to penetrate through the calyx area (preferred site of entry), and through the side of the fruit. Lower success to penetrate through the side of the fruit could be related to the presence of some physical and/or chemical barrier in the fruit epidermis of 'Anjou' with antixenotic and/or antibiotic effects on neonate larvae. The same factors could be present in fruits of both pear cultivars at the beginning of the season, when penetration through the side of fruit was low in pears. At the end of the season, the calyx

area was not the preferred penetration site. Therefore, the lower difference on the penetration success in 'Bartlett' and 'Anjou' pears was probably related to the decreasing difference on the ability to penetrate through the free surface of these two host fruits. These results and observations suggest that physical, anatomical and, probably, chemical factors could be responsible for some degree of resistance in less susceptible host cultivars, such as 'Anjou'.

Westigard et al. (1976) explained the different susceptibility to larval entry between apple and different pear cultivars in terms of physical barriers associated with lignification (stone cell formation). Suomi et al. (1986) reported that some chloroform extractable materials from several plants (such as rabbitbrush, *Chrysothamnus nauseosus* (Pallas), wormwood, *Artemisia absinthium* (L.), and tansy, *Tanacetum vulgare* (L.)) could be possible feeding deterrents for neonate codling moth larvae. More detailed studies (i.e. analyses with electron microscopy) should be conducted to determine the physical and/or chemical factors in the epidermis of 'Anjou' fruit responsible for resistance to larval penetration. This would help to a better understanding of the basis of the resistance to codling moth attack at the beginning and middle of the season in winter pear cultivars.

Mortality of larvae once established inside the fruit was similar in apples and pears. Therefore, it does not account for the differences in infestation levels observed in the field among apple cultivars. At the beginning and end of the season, larval mortality inside the fruit was higher than in mid-season. The higher incidence of larval mortality early in the season could be associated to the fall of fruit (June fall). After a fruit falls, it starts to dehydrate, faster on pear than on apple fruits, and larvae probably do not find the appropriate conditions to develop. Similar inappropriate conditions seemed to exist when fruit approached or reached maturity. Mortality of established larvae inside the fruit was similar to that on unsprayed 'Gravenstein' apple as reported by Wood (1965) who found 11% mortality after larval penetration. Hagley (1972b) observed that mortality of codling moth larvae inside apple fruit was highly correlated with total rainfall and affected mostly first instar larvae when they were just beneath the epidermis of the fruit. Knight (1998) reported similar results from field tests with overhead watering. In our studies, mortality

inside the fruit was not associated with rainfall because there was no rain during the days the experiments were conducted.

The decrease in penetration success when larvae were starved for 8 h was especially important in pear cultivars. This could be due to a greater skin and flesh firmness in pear compared with apple fruit at the time of the season when the experiments were conducted. Fruit with greater skin firmness would require more energy to penetrate. Moreover, larval movement was more restricted and larvae tried to penetrate near the point where they were released (petiole area). Therefore most larvae were unable to reach the calyx area, that was still the preferred penetration site in 'Bartlett' pears at the time of the season when the experiments were conducted. Cutright (1931) suggested that short starvation time (i.e., 2 h) could stimulate larval activity, which, in turn, would increase the likelihood of larval penetration. However, Jackson & Harwood (1980) observed an inverse linear relationship between starvation time and penetration success in apple.

5.5 Conclusions

There are not differences in natural mortality of the codling moth in different host cultivars. Therefore, differential susceptibility to codling moth attack among host cultivars is not associated to natural mortality of the egg stage. Moreover, duration of the development of the egg stage is also similar in different oviposition substrates of apple and pear cultivars.

Travel speed of neonate larvae when searching for a fruit is similar in different cultivars. Moreover, average distance of eggs to the nearest fruit is not significantly different among hosts. Consequently, no important differences in neonate mortality before fruit penetration should be expected among cultivars.

There are significant differences in first instar larval mortality among host cultivars over most of the season. Mortality is higher on the 'Anjou' winter pear cultivar than on the 'Bartlett' summer cultivar and 'Red Delicious' apple, except at the end of the season.

Differential first instar mortality is due to the different success to enter the fruit. The ability to penetrate the fruit is correlated with infestation levels observed in the field. Therefore, first instar larval mortality while attempting to enter fruit is the key factor which explains differences in susceptibility to codling moth attack among host cultivars.

The existence of resistance to first instar larval penetration into the fruit of 'Anjou' pears, as well as other winter pear cultivars, during most of the season should be used to develop cultivar-specific control programs. Higher treatment and economic injury levels should be defined for these cultivars during that period of the season they show resistance to larval penetration. The implementation of such thresholds will reduce the need of chemical intervention, decreasing non-desirable effects of chemicals in the environment as well as on the public health (including farm workers) and natural enemies of pests.

The ability to penetrate the fruit is related to neonate behavior on the fruit surface and to physical, anatomical, and chemical characteristics of host fruits. At the beginning of the season, once on the fruit surface, neonate larvae moved directly to the calyx area. This is the preferred fruit site where they attempt to enter into the fruit. Therefore, characteristics of the fruit in that area are very important to explain the ability of neonate larvae to penetrate. Apple fruits have a lower skin firmness except when pears approach maturity and lack a layer of stone cells during the whole season. This explains why neonate larvae are successful penetrating apple fruits during the whole season. Pear fruits have a layer of stone cells that represents a physical barrier for neonate larvae to enter the fruit. The layer of stone cells is continuous in 'Anjou' pears. However, it has a discontinuity in the calyx area in 'Bartlett' pears. This discontinuity acts as a 'channel' through which neonate larvae can penetrate easily during the whole season. At the middle of the season, neonate larvae attempt to penetrate both through the calyx area and the side of the fruit. The presence of some physical and/or chemical barrier in the fruit epidermis of 'Anjou' pears with antixenotic and/or antibiotic effects on neonate larvae are likely responsible for the differences observed in penetration success of neonates at the middle of the season. More studies and fruit analyses are needed to try to determine what these factors are and how they affect larval performance. When pears are close to maturity the percentage of neonate larvae successfully entering the fruit is similar in different apple and pear cultivars.

The understanding of the basis of such resistance to larval penetration can be the starting point to develop resistant material that can be used effectively in commercial orchards as an alternative to management tactics currently used.

Larval mortality inside the fruit does not account for the differences in infestation levels observed in the field among apple cultivars.

CHAPTER 6

**EFFECT OF FOOD SOURCE
ON LARVAL AND PUPAL DEVELOPMENT, ADULT FECUNDITY
AND EGG VIABILITY IN DIFFERENT CODLING MOTH
(LEPIDOPTERA: TORTRICIDAE) STRAINS**

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6.1 Introduction

The codling moth, *Cydia pomonella* L. (Lepidoptera: Tortricidae) is an important worldwide pest of deciduous tree fruits. Pome fruits (apples and pears) are the preferred hosts (Shel'Deshova 1967, Riedl 1983) but certain stone fruits, such as apricots (Madsen & Borden 1954), and walnuts (Quayle 1926, Putman 1963) are also attacked. Phillips & Barnes (1974) inferred from studies with host races from apple, plum and walnut in California that they differed not only in terms of developmental heat requirements but also in terms of behavioral traits. Cisneros & Barnes (1974) found that host races may also differ in terms of fecundity, the total number of eggs produced by a female. These differences appear to have a strong genetic basis, but, in addition, host specific nutritional factors may also contribute to differences in developmental, behavioral and reproductive traits between host races.

Food quality can have a pronounced effect on larval development. For instance, larvae of the polyphagous obliquebanded leafroller, *Choristoneura rosaceana* Harris (Lepidoptera: Tortricidae) developed faster when reared on young than on old apple foliage (Onstad & Reissig 1986). However, development was similar on leaves of different apple cultivars (Onstad & Reissig 1986).

In the codling moth, larval development as a function of temperature has been studied in semi-field conditions or in the laboratory with apples or various artificial diets as food source (Glenn 1922, Rock & Shaffer 1983, Setyobudi 1989). These data have been used to calculate physiological thresholds and heat unit requirements (degree-days) for the immature stages of the codling moth. This information is used in pest management to predict codling moth phenology for improved timing of control measures (Riedl & Croft 1978, Brunner & Hoyt 1982). Similar data are lacking for pears. It cannot be assumed that larval development on pears is the same as on apples since pears represent a different larval medium which may have unique nutritional characteristics. Larval nutrition may also affect the reproductive potential of codling moth on a particular host in terms of fecundity and egg viability.

This study examines the effect of food quality (apple vs. pear) on larval and pupal development, fecundity (total number of eggs produced/female) and egg viability in a laboratory strain and two field strains (one from apple, one from pear). The purpose of this research was to develop host-specific information on codling moth development and examine nutritional factors which might determine the reproductive success of codling moth on apple and pears.

6.2 Materials and Methods

Codling moth populations. A population of *C. pomonella*, collected in 1991 in an apple block at the Mid-Columbia Agricultural Research and Extension Center (MCAREC), Hood River, OR, was maintained on thinning apples in an insectary at 24 ± 2.5 °C, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h, as described by Riedl & Loher (1980). This laboratory population supplied eggs for larval development studies and adults for fecundity and egg viability tests. In addition, adults which developed from diapausing larvae collected in the field during the summer of 1999 from ‘Red Delicious’ apple (MCAREC), and ‘Anjou’ and ‘Bartlett’ pear (Southern Oregon Experiment and Extension Center, Medford, OR), were used for fecundity and egg viability comparisons.

Research sites and plant material. One apple, ‘Red Delicious’, and two pear cultivars, ‘Anjou’ and ‘Bartlett’, were used for larval and adult tests. Unsprayed plant material was used in all experiments. Larval development experiments were conducted in the field in an apple and in a mixed pear block located at the MCAREC. The apple block was 0.5 ha in size with a tree and row spacing of 6.1 m. It consisted of alternating rows of 31-year-old ‘Red Delicious’ and ‘Newtown’ apple trees. The 0.2-ha pear block consisted originally of alternating rows of 35-year old ‘Anjou’ and ‘Bartlett’ pear trees with a tree and row spacing of 8 m. Adult fecundity experiments were conducted in temperature-controlled limb cages (Mellenthin & Bonney 1972) located next to an apple and a pear block at the MCAREC.

Effect of food source on larval and pupal development. The hypothesis was tested that the duration of larval and pupal development depends on the food source. The experiment was conducted in the field with 'Red Delicious' apples, and 'Anjou' and 'Bartlett' pears. Individual fruits were enclosed in organandy bags (10 cm diameter x 15 cm long) protect eggs from predation and prevent fruit from being attacked by other pests. A piece of wax paper (ca. 10 x 10 mm) with a single codling moth egg in the 'black head' stage of development was attached with Elmer's Glue (Elmer's Products, Inc. Columbus, OH) to each fruit next to the petiole area. A piece of 2 x 3 cm of corrugated cardboard was placed inside the bag to provide a pupation site for larvae after they left the fruit. The experiment was repeated three times (May to July) during the 1998 season. One hundred eggs for each cultivar were used each time. Fruit was checked daily, and records were kept of the number of eggs hatched, successful entries, and larval survivorship inside the fruit. Development of codling moth individuals was followed until completion of larval development and pupation.

Effect of larval food source on adult fecundity and egg viability. The hypothesis was tested that food source (apple or pear) has no effect on egg production and egg viability. Experiments were conducted in semi-field conditions in portable limb cages (Mellenthin & Bonney 1972) under fluctuating temperature (ranging from 21 to 29 °C), $75 \pm 5\%$ RH, and natural light supplemented by an artificial photoperiod of 17:7 (L:D) h. Tests were conducted with laboratory and field moths. Laboratory moths were used in tests with all host fruits. The field strains were only used in experiments with fruit of the host cultivar on which they were collected. First generation field moths were collected in the field as larvae and spent the winter in diapause. Second generation field moths were reared separately on fruit of the corresponding cultivar in the insectary at $24 \pm 2.5^\circ\text{C}$, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h. One pair of virgin moths was placed inside a 350 ml wax paper cup for a 7-day period (100 degree-days). Moths were able to perceive visual and olfactory stimuli from a 'Red Delicious' apple (ca. 30 mm diameter) placed on a site of the cup. Adults were fed with water and a solution of 5% sucrose (supplied from cotton wicks). Under these conditions, the majority of moths (> 90%) mated the first night and adult females laid most of the eggs during the 7-day time period. Tests were conducted over two

consecutive generations. About twenty replications of one adult pair each were carried out for each host cultivar and generation. Moths laid eggs on the interior walls of the wax paper cups. Egg laying between field and laboratory moths reared on different host fruits was compared. Only mated adult pairs which produced eggs were included in the analyses. Cups with eggs were placed in an insectary at 24 ± 2.5 °C, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h. Egg development was checked and egg viability (expressed in terms of egg survivorship) was recorded.

Measurement of environmental variables and calculation of degree-days (DD). In all experiments, temperature and relative humidity measurements were taken with HOBO and StowAway loggers (Onset Instruments, Pocasset, MA, U.S.A.). Temperature (both inside and outside the fruit) and relative humidity were recorded. Degree-day requirements for completion of larval and pupal development in different apple and pear cultivars were compared using lower and upper thresholds of 10 and 31 °C, respectively (Glenn 1922). Hourly temperatures were recorded by inserting a thermistor sensor in the side of the fruit. Degree-days for larval development were based on internal fruit temperatures and were calculated by accumulating degree-hours over each 24-h period. Hourly temperatures outside the fruit were used to calculate degree-days for pupal development.

Statistical Analysis. Analysis of variance (ANOVA) and contrasts for mean comparison according to Waller-Duncan Bayes LSD were carried out with the Number Cruncher Statistical System (Hintze 1997) to compare heat unit requirements (degree-days) of larvae and pupae as well as fecundity of individuals reared on different food sources. Fecundity data were transformed to $\log(x+1)$ for the analysis. Untransformed data are reported in the text and tables. Analyses of developmental rates were supported by survival analysis tests (Proportional Hazard Regression, PHREG; SAS 1997). Logistic regression for binomial responses (SAS 1997) was used to analyze egg viability data.

6.3 Results

Effect of food source on larval and pupal development. The average duration of larval development of the MCAREC laboratory strain ranged from 276.8 to 287.3 degree-days (Table 6.1). There were no differences in the duration of larval development on different host cultivars ($F = 2.62$; $df = 2, 224$; $P = 0.075$; Table 6.1). Maturity stage of the fruit did not affect larval development rate ($F = 0.03$; $df = 2, 224$; $P = 0.974$; Table 6.1). Duration of larval development was statistically different among sexes ($F = 7.82$; $df = 1, 224$; $P = 0.006$) when sex was the only factor considered in the analysis and all individuals were considered together. Females required about 5-10 degree days more to complete larval development. However, heat requirements for completion of larval development were not significantly different among sexes when in addition to sex two other factors, cultivar and time of year, were considered in the analysis (Table 6.1). Interactions were not significant.

Table 6.1: Thermal requirements for the development of the larval stage of the MCAREC laboratory codling moth strain. Means for male and female larvae for each cultivar and time of year, and combined means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns = not significant.

Time of year	Cultivar	Larval development (DD)					
		Male		Female		Combined	
		n	Mean (SEM)	n	Mean (SEM)	n	Mean (SEM)
1-Jun	Red Delicious	16	272.2 (7.7) ns	16	281.4 (7.7)	32	276.8 (5.4) ns
	Anjou	6	273.2 (8.7) ns	7	298.1 (8.8)	13	286.6 (6.9)
	Bartlett	24	280.4 (4.7) ns	19	283.0 (5.4)	43	281.5 (3.5)
23-Jun	Red Delicious	10	269.8 (6.6) ns	11	285.7 (6.0)	21	278.1 (4.7)
	Anjou	5	283.1 (11.4) ns	3	292.6 (16.0)	8	286.7 (8.8)
	Bartlett	13	276.9 (5.7) ns	17	281.8 (4.7)	30	279.9 (3.6)
15-Jul	Red Delicious	21	275.9 (4.2) ns	31	282.2 (3.4)	52	279.6 (2.7)
	Anjou	8	284.4 (4.9) ns	7	290.7 (7.8)	15	287.3 (4.4)
	Bartlett	9	279.3 (6.1) ns	19	281.2 (4.3)	28	280.6 (3.4)

Average duration of pupal development of the MCAREC laboratory strain ranged from 167.7 to 171.6 degree-days (Table 6.2). No differences in the duration of the development of the pupal stage were observed among different host cultivars ($F = 1.63$; $df = 2, 224$; $P = 0.198$). Stage of maturity of the fruit did not affect duration of pupal development ($F = 1.51$; $df = 2, 224$; $P = 0.223$). Duration of pupal development was statistically different among sexes ($F = 15.48$; $df = 1, 224$; $P < 0.001$) when sex was the only factor considered in the analysis and all individuals were considered together. Females required about 5 degree-days more to complete pupal development. However, heat requirements for completion of pupal development were not significantly different among sexes when in addition to sex two other factors, cultivar and time of year, were considered in the analysis (Table 6.2). Interactions were not significant.

Table 6.2: Thermal requirements for the development of the pupal stage of the MCAREC laboratory codling moth strain. Means for male and female pupae for each cultivar and time of year, and combined means not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$); ns: not significant.

Time of year	Cultivar	Pupal development (DD)					
		Male		Female		Combined	
		n	Mean (SEM)	n	Mean (SEM)	n	Mean (SEM)
1-Jun	Red Delicious	16	170.0 (1.6) ns	16	171.6 (2.3)	32	170.8 (1.4) ns
	Anjou	6	168.1 (3.8) ns	7	174.6 (3.6)	13	171.6 (2.7)
	Bartlett	24	166.5 (1.7) ns	19	171.1 (2.0)	43	168.7 (1.3)
23-Jun	Red Delicious	10	171.7 (1.4) ns	11	168.2 (1.7)	21	169.8 (1.2)
	Anjou	5	169.0 (1.3) ns	3	172.8 (4.1)	8	170.4 (1.7)
	Bartlett	13	165.8 (2.4) ns	17	170.4 (1.7)	30	168.4 (1.4)
15-Jul	Red Delicious	21	162.1 (1.3) ns	31	171.4 (1.1)	52	167.7 (1.1)
	Anjou	8	166.8 (3.7) ns	7	172.6 (2.3)	15	169.5 (2.3)
	Bartlett	9	165.1 (1.8) ns	19	170.4 (1.3)	28	168.7 (1.1)

Effect of larval food source on adult fecundity and egg viability. Average adult fecundity ranged from 116.9 to 143.4 eggs/female (Table 6.3). Larval food did not significantly affect adult fecundity ($F = 0.66$; $df = 2, 205$; $P = 0.518$; Table 6.3). No significant differences in fecundity were observed among laboratory and field strains ($F = 0.73$; $df = 1, 205$; $P = 0.730$). Average fecundities of the overwintering generation (ranging from 116.8 to 127.0 eggs/female) tended to be lower, although not significantly ($F = 3.50$; $df = 1, 215$; $P = 0.063$), than average fecundities of the summer generation of field strains (ranging from 131.9 to 143.4 eggs/female).

Viability of eggs, expressed in terms of egg survivorship, laid by adults reared on different food sources ranged from 91.0 to 94.7 %. Larval food source did not significantly affect egg viability of individuals of the next generation ($\chi^2 = 0.0095$; $df = 2$; $P = 0.920$; from regression analysis for binomial responses). There were no significant differences on egg viability among codling moth strains ($\chi^2 = 2.330$; $df = 1$; $P = 0.127$; from regression analysis for binomial responses) and generations ($\chi^2 = 1.876$; $df = 1$; $P = 0.171$, from regression analysis for binomial responses). No interactions were significant.

Table 6.3: Adult fecundity and egg viability (survivorship) of laboratory and field codling moth strains reared on different host fruits. Mean adult female fecundities and weights not followed by the same letter are significantly different according to Waller-Duncan Bayes LSD ($\alpha = 0.05$). Egg survivorship data not followed by the same letter are significantly different according to logistic regression for binomial responses; ns: not significant.

Strain	Cultivar	Generation	n	Weight (mg)	Fecundity (eggs/female)		Egg survivorship (%)
					Mean (SEM)	Range	
Lab	Red Delicious	n	20	21.8 ns	139.7 (11.2) ns	(56-229)	91.2 ns
	Anjou	1st	20	21.2	128.3 (15.9)	(29-256)	91.5
	Bartlett	1st	20	21.0	131.3 (17.0)	(8-242)	91.9
	Red Delicious	n+1	20	21.3	140.6 (13.8)	(23-235)	91.0
	Anjou	2nd	20	20.9	134.9 (14.7)	(34-214)	92.0
	Bartlett	2nd	20	20.9	134.9 (16.2)	(36-272)	90.2
	Red Delicious	n (winter)	19	21.2	127.0 (15.9)	(5-216)	93.2
	Anjou	n (winter)	8	19.7	118.4 (24.6)	(19-177)	92.6
	Bartlett	n (winter)	19	19.7	116.9 (16.1)	(3-260)	94.7
Field	Red Delicious	n+1 (summer)	20	21.7	143.4 (13.7)	(62-259)	91.0
	Anjou	n+1 (summer)	14	20.9	131.9 (14.8)	(29-223)	91.0
	Bartlett	n+1 (summer)	17	20.9	134.8 (13.4)	(58-261)	91.3

6.4 Discussion

The average heat requirement for completion of larval development ranged from 276.8 to 287.3 degree-days. These heat unit totals were calculated from temperature measurements taken inside fruit and were similar to those reported by other authors who used ambient air temperatures. Setyobudi (1989) found that the thermal requirement for larval development was 288.9 degree-days (lower threshold = 11.2; upper threshold no reported). Pitcairn et al. (1991) reported that 280 degree-days were required to complete larval development (lower threshold = 10 °C; upper threshold = 32 °C). Martí (1995) found that 305 degree-days (lower threshold = 11 °C; upper threshold = 32 °C) were required to complete larval development of a Spanish population. Differences on heat requirements reported by authors can be related to the use of different development thresholds, to actual different requirements among strains, and/or to the larval food source. Setyobudi (1989), Pitcairn et al. (1991), and Martí (1995) worked with laboratory strains. Martí (1995) studied development in apple fruit in semi-field conditions. Setyobudi (1989) and Pitcairn et al. (1991) conducted their studies with artificial diet in the laboratory.

The data showed the trend that larval development might be slower in ‘Anjou’ pears than in ‘Red Delicious’ apple and ‘Bartlett’ pear. Individuals fed on ‘Anjou’ pears required about 5-10 degree-days more to complete larval development. The development of fewer individuals was followed in ‘Anjou’ pears. This was related to the fact that penetration success in ‘Anjou’ pears is significantly lower than in apple and ‘Bartlett’ pears. A higher proportion of first-instar larvae died while trying fruit penetration in ‘Anjou’ pears. This observed trend could become statistically significant with more data on larval development in ‘Anjou’ pear fruits. This should be confirmed in subsequent studies. The observed tendency that higher heat requirement for larval development is required in ‘Anjou’ pears could also be related to a longer time spent by first-instar larvae trying to penetrate the fruit. Two days after the eggs in the ‘black head’ stage were attached to the fruit, larvae were seen wandering on the surface of ‘Anjou’ pears. At that time, larvae already had penetrate into ‘Red Delicious’ apples and ‘Bartlett’ pears. Differential developmental rates could also be associated to differences on food quality among cultivars. Phillips & Barnes (1975)

determined that a plum strain required less heat than an apple or walnut strain to complete development. These differences appeared to have a strong genetic basis, but, in addition, host specific nutritional factors might also contribute to differences in developmental rates between host races.

Data suggest that the development stage of the fruit does not affect larval developmental rate. In contrast to these results, several authors have observed faster larval development in immature fruits (Hall 1934, Gentner 1940, Marshall 1940, Heriot & Waddell 1942, and Cisneros & Barnes 1974). Heriot & Waddell (1942) suggested that immature tissues have a greater nutritive value than mature fruit tissue, both seeds and pulp, and that was the reason why larval development was faster in immature fruits. However, Isley (1939) reported that the degree of maturity of the fruit had no influence on the duration of larval development.

Although females required about 5-10 degree days more to complete larval development, these differences are not important biologically and for management decision making. During one day more than 10 degree-days can accumulate, especially in days with moderate and high temperatures.

The average duration of pupal development ranged from 167.7 to 171.6 degree-days which falls in the range of heat requirements for pupae reported by other authors. Setyobudi (1989) found that the thermal requirement for pupal development 152.3 degree-days (lower threshold = 9.8; upper threshold no reported). Pitcairn et al. (1991) reported that 260 degree-days were required for completion of pupal development (lower threshold = 10 °C; upper threshold = 28 °C). Martí (1995) found that 148 degree-days (lower threshold = 11 °C; upper threshold = 32 °C) were required to complete pupal development of a Spanish population. Differences on heat requirements can be related to the use of different development thresholds, to actual different requirements among strains, and/or to the larval food source.

Results suggest that neither food source nor the stage of fruit maturity affect the development of the pupal stage. However, analyses suggest that duration of pupal development is different among sexes when only the sex factor is considered. Females required on average 5 degree-days more to complete pupal development. However,

differences among sexes are not important biologically and for management decision making, since 10 degree-days can easily accumulate in one day, especially in summer.

The average degree-days from newly emerged larvae to adults ranged from 447 to 458 degree-days, values lower compared to other studies (all base 10 °C): 552.8 degree-days (Glenn 1922), and 514 degree-days (Rock & Shaffer 1983).

Results suggest that codling moth larval and pupal development are not significantly different among apple and pear cultivars. Therefore, phenology models developed from data with apple as food source seem to be valid also for pear. This especially applies when apple and pear trees or orchards are mixed. In this situation may be migration from contiguous trees or orchards of different cultivars. Therefore there is gene flow that avoids the formation of host races. In fact, the laboratory strain at the MCAREC was collected from apples, but apple and pear blocks are mixed at the MCAREC. However, the possibility exists that geographic isolation and adaptation to pear occurs in areas where there is monoculture of pear cultivars for a long time. Codling moth apple and pear races could indeed have different larval and pupal development requirements.

Results also indicate that larval food source does not affect adult fecundity and egg viability (survivorship) of the next generation. Deseö (1971) reported that the quality and quantity of larval nutrition influence the fecundity of codling moth females. Cisneros & Barnes (1974) found that a walnut population had a higher reproductive potential than an apple population and related the differences in fecundity to differences in the larval medium. The nutritive value of apple and pear for codling moth larval development and reproduction seem to be similar since there were not differences in adult weight, fecundity and egg viability. One can conclude from these results that differences in susceptibility to codling moth among apple and pear cultivars is not a manifestation of differential adult fecundity.

The average fecundities of field female moths ranged from 116.9 to 143.4 eggs/female, and were higher than the previously reported estimates. Hall (1929) obtained an average of 83 eggs/female and a maximum of 208 eggs/female from first brood moths at

Vineland Station, Ontario, Canada. Geier (1963) reported a much lower mean of 44 and a maximum of 198 eggs/female in the Capital Territory of Australia. In our experiments, adults were fed with water and a 5% sucrose solution which may have increased fecundity. Wiesmann (1935) obtained twice as many eggs from sugar-fed females as from unfed individuals. Hamstead & Gould (1950) suggested that starvation causes females to re-absorb part of their maturing oocytes. The average fecundity of laboratory moths reared on different host fruits ranged from 128.3 to 140.6 eggs/female and were lower than the one reported by Gehring & Madsen (1963), who found means over an 8-day period of 179.1 and 162.5 eggs/female for two different strains.

Field confined moths, in a density of 1 male: 1 female, oviposited regularly. This is in contrast to the finding of many authors, including Putman (1963) and it has an important implication for the studies conducted with laboratory reared moths. Successive generations of moths reared under semi-field conditions are expected to preserve the genetic variability present in the field strain. Codling moth strains reared in laboratory conditions have been found to lose the genetic variability. This can affect the behavior and biological parameters (such as fecundity, development) of the individuals of the laboratory reared strains. This suggests that strains kept for experimental purposes should be reared in semi-field conditions in order to get more reliable results.

Average fecundities of the overwintering generation ranged from 116.8 to 127.0 eggs/female and tended to be lower than average fecundities of the spring generation of field strains that ranged from 131.9 to 143.4 eggs/female. Trottier & Hagley (1979) reported that the average fecundity over several years varied from 9 to 30 eggs/female of the overwintering generation of a codling moth strain from Ontario, Canada. Wearing & Ferguson (1971), Deseö (1973), and Deseö & Saringer (1975a, 1975b) also noted differences in fecundity among codling moth generations. Fecundity was lower (17-23 eggs/female) in individuals of the overwintering generation than in individuals of the first generation (76-83 eggs/female). Geier (1963) related low fecundity in females from the overwintering generation to depletion of body reserves during winter diapause.

6.5 Conclusions

Codling moth larval and pupal development rates are not significantly different in apple and pear fruits. This suggests that phenology models of the codling moth developed from data with apple as food source are valid for pears. However, there is the possibility that long time geographic isolation and adaptation to pear host occurs. In this case, codling moth apple and pear races could indeed have different larval and pupal development requirements. This is not likely to occur when apple and pear orchards are mixed. In this situation, movement of adult moths between contiguous orchards and, consequently, gene flow of individuals reared on different cultivars is more likely to occur.

Adult fecundity and egg viability of different apple and pear codling moth strains is not different. Therefore, these biological parameters are not correlated to differences in susceptibility to codling moth attack among apple and pear cultivars.

CHAPTER 7

SUMMARY AND CONCLUSIONS

This study was conducted to investigate several key aspects of the relationship between the codling moth, *Cydia pomonella* L. (Lepidoptera: Tortricidae) and its principal hosts, apple and pear.

One main objective was to investigate the responses of the adult stage of the codling moth to host (apple and pear) and non-host (maple) trees and to determine the role of visual and olfactory cues in guiding adults to host plants. In release experiments, codling moth adults were intercepted in significantly greater numbers in host (apple, pear) than in non-host (maple) trees. These observations suggest that codling moth adults exhibit directed movement in response to cues from host plants and do not orient to and select host plants randomly. However, there was no correlation between adult preference for certain hosts and their susceptibility to codling moth attack. 'Bartlett' and 'Anjou' pear trees were consistently more attractive to the adult stage than 'Newtown' apple trees. However, in other experiments 'Red Delicious' apple and 'Bartlett' pear were similarly attractive. These observations suggest that host preferences by adult moths do not explain differences in susceptibility to codling moth attack among host cultivars. Cues from the foliage of host trees were important in orienting codling moth adults to the host plant during the whole season. Olfactory and visual cues from host fruit provided additional stimuli to adults in host selection, except at the beginning of the season when fruit was small.

These results showed that the relative importance of visual and olfactory stimuli from the canopy and fruit in guiding codling moth adults to a plant is dependent on the distance to the source. At short distances (5, 10 m) olfactory stimuli from foliage and visual and olfactory cues from fruit seemed to be key factors for host selection. At greater distances (15 m) moths were mostly guided by olfactory cues from host fruit.

Alpha-farnesene is a volatile sesquiterpene released by apple and pear fruits, and reportedly affects oviposition behavior of female moths and searching behavior of neonate larvae. Leaves and fruits of pear cultivars had a higher content of α -farnesene than leaves and fruits of 'Red Delicious' apple. Among pear cultivars, α -farnesene content was higher in 'Anjou' than in 'Bartlett' pear. Results showed no correlation between the preferences moths displayed for the two pear cultivars 'Anjou' and 'Bartlett' and α -farnesene content in leaves.

Canopy and fruit shape did not affect adult response to host trees. However, experiments indicated that fruit size and color are visual cues perceived by codling moth adults.

Results showed that different codling moth strains have similar behavior patterns during host selection. However, moths reared on apples were guided more by olfactory cues from fruit than sterilized moths reared on diet. These observations suggest that sterile moths, in spite of years of laboratory rearing and radiation treatment with Cobalt-60, behave similarly to non-sterile moths reared on apples and still respond to and recognize host trees. This is important, since the fruit industry in British Columbia, Canada, is presently engaged in a multi-year sterile insect release (SIR) program to control codling moth in a limited geographic area.

The second objective of this study was to determine how adult mating and oviposition behaviors varied among host trees and how these behaviors related to differences in susceptibility. Results showed that under caged conditions codling moth is able to mate in non-host trees. However, codling moth mating rate was higher in host plants than in non-host plants. In host plants, stimuli from fruit were not essential for mating. Stimuli from foliage of host cultivars seemed to be enough to trigger mating. However, in non-host plants, host fruit had a positive effect both on the percentage of mated females and oviposition activity. Codling moth mating rate was similar in different host cultivars, including 'Red Delicious' apple and 'Anjou' and 'Bartlett' pears. The egg distribution in the field varied through the season and with the cultivar. However, mean distance of eggs to the nearest fruit were not significantly different among cultivars. Egg distribution was random with respect to individual fruits, but not with respect to fruit clusters of different

fruit density. Feral and laboratory female moths did not discriminate among clusters with non-infested and codling moth infested fruit and accepted both equally as oviposition sites. However, in the field no egg laying occurred on clusters with ripe 'Bartlett' pears. This behavior may be related to the presence of ethylene, a volatile produced that is released by 'Bartlett' pears when they reach maturity. Ethylene, in the concentrations found in the intercellular spaces of ripe 'Bartlett' pears, adversely affected codling moth mating and it tended to decrease egg laying. No egg sampling was conducted at the time that 'Red Delicious' apple and 'Anjou' pear ripened on the tree. Differences in egg distribution should be taken into account when selecting insecticides for greatest effectiveness. Some insecticides currently used against codling moth eggs, such as some mineral oils, have been shown to lead to a different degree of control when applied on different oviposition substrates (fruit and leaves) (Riedl et al. 1998).

Additional multiple-choice experiments showed that selection of an oviposition site is affected by anatomical characteristics (e.g., pubescence) of oviposition substrates. Pubescent leaf surfaces (e.g., underside of 'Red Delicious' apple leaves) were avoided as oviposition sites. Smooth leaves of non-host species (maple) had no particular attraction for oviposition. This was probably related to chemical factors, such as lower content of oviposition stimulants (α -farnesene). Oviposition deterrents could also be involved. However, other factors such as the position of the leaf surface, the orientation of the leaf surface relative to a fruit and to a light source, and the distance to a fruit, were also important and affected the location where female moths placed eggs.

The third objective was to determine whether or not differences in codling moth infestation levels are a manifestation of differential mortality of immature codling moth stages on different host fruits. Results showed not differences in natural egg mortality (about 10%) in different host cultivars ('Red Delicious' apple, 'Anjou' and 'Bartlett' pear). Therefore, differential susceptibility to codling moth is not associated with mortality of the egg stage. The duration of development of the egg stage was also similar on different oviposition substrates of apple and pear. Travel speed of neonate larvae when searching for a fruit was also similar in different cultivars. The average distance of egg to the nearest fruit was not significantly different among hosts. Therefore, no significant differences in

neonate mortality while searching for the food source (fruit) should be expected among host cultivars.

However, there were significant differences in first instar larval mortality among host cultivars over most of the season. Mortality was much higher on the 'Anjou' winter pear than on the 'Bartlett' summer pear and 'Red Delicious' apple, except at the end of the season. Differential first instar mortality was due to the different success rates in entering the fruit. The ability to penetrate the fruit was correlated with infestation levels observed in the field and was related both to neonate behavior and anatomical characteristics (stone cells) of host fruits.

At the beginning of the season, neonate larvae placed on the fruit surface oriented mostly to the calyx area both on apple and pear. This was the preferred site of the fruit to attempt penetration. Characteristics of fruit anatomy in the calyx area are very important to explain the penetration ability of neonate larvae. Skin firmness in apple is lower than in pear during most of the season. Apples lack a layer of stone cells. Pear fruits have a layer of stone cells that represents a physical barrier for neonate larvae to enter the fruit. The layer of stone cells is continuous in 'Anjou' pears. In 'Bartlett', the area just below the calyx is free of stone cells which allows neonate larvae to penetrate to the fruit interior. At the middle of the season, neonate larvae tried to penetrate mostly through the side of the fruit. The presence of a barrier in the fruit epidermis of 'Anjou' pears with antixenotic and/or antibiotic effects on neonate larvae was likely responsible for the differences observed in neonate penetration rates at the middle of the season. More studies and fruit analyses are needed to determine what these factors are and how they affect neonate larval performance. When pears were close to maturity the percentage of neonate larvae successfully entering the fruit was similar in different cultivars. Once larvae had entered the fruit, there were no differences in larval mortality among cultivars. Pupal mortality was similar among host cultivars during the whole season.

The last main objective was to determine the effect of the food source (host fruit) on codling moth post-embryonic development rates and adult reproductive capacity. Results showed that larval food source did not affect larval and pupal developmental rates, adult fecundity, and viability of eggs of the next generation.

The results of this study suggest that mortality of first instar larvae while attempting to enter fruit is the key factor explaining differences in susceptibility to codling moth attack among host cultivars. Differences in susceptibility to codling moth attack among cultivars can be exploited to develop cultivar-specific control programs. Higher treatment and economic injury levels can be used for cultivars that show resistance to larval penetration. The implementation of such thresholds will reduce the need for chemical intervention. Moreover, this strategy is compatible with the implementation of softer control programs.

The results of the studies conducted in each chapter can be combined and used to generate a simple life table. The construction of a life table is an important component in the understanding of the population dynamics of an insect species. The life table would be a useful tool to estimate the variations in population size, from generation to generation, and, consequently, the number of viable individuals at the end of the season, which would represent the fitness of the codling moth in each cultivar. The number of viable individuals in each host cultivar would be correlated with the infestation levels observed in the field. In addition, the life table would provide information about the role of different factors affecting the population dynamics of the species.

An example of the use of the life table is presented in Table 7.1. This example represents an imaginary situation with a mixed block having trees of three host cultivars: 'Red Delicious' apple, 'Anjou' and 'Bartlett' pears. To make the case simpler, the assumptions are that (1) the block contains the same number of trees of each cultivar (randomly distributed), (2) trees of different cultivars are of the same size (volume, height, width) and have the same fruit density, (3) there are two codling moth generations per year in the area where the block is located, (4) there is no initial population of codling moth in the orchard, (5) there is migration of adult codling moths (e.g., 20 pairs) into the block only once at the beginning of the season, and (6) there is no chemical intervention against the codling moth and no control of the pest by natural enemies.

The number of successful first-instar larvae penetrating the fruit gives an idea of the number of infested fruit in different host cultivars, assuming that only one fruit is attacked by one larva. Estimates of the number of first-instar larvae causing stings (shallow feedings) could also be included to predict the level of damaged fruit.

Data of survivorship of inside the fruit as well as survivorship of overwintering larvae in different host cultivars would help to get estimates of potential population levels for next year.

Variables such as number of trees, size and fruit density of trees can indeed not be the same for each cultivar. This would affect the number of migrating moths colonizing trees of each cultivar. Therefore, these variables should be considered in the model by changing the value of the estimate of the percentage of moths colonizing trees of each cultivar.

The research followed the codling moth's life cycle and focused on how factors associated to different host cultivars affected some aspects of the biology and behavior of the codling moth. The life table presented here has the same structure and summarizes the success of the codling moth to reproduce and survive on apple and pear cultivars.

The percentage of moths selecting hosts in Table 7.1 is the estimate of the proportion of the total number of migrating adults which would find and select each specific host cultivar. The values are estimated from the release experiments conducted with sterile moths (Chapter 3). Feral moths, as well as moths of different host races, could show different behaviors. Since there was no preference for 'Red Delicious' or 'Bartlett' trees in the release experiments conducted at The Dalles site, but consistently slightly more adults were recaptured in 'Bartlett' than in 'Anjou' trees at the MCAREC site, the estimates are 35, 30, and 35% for 'Red Delicious', 'Anjou', and 'Bartlett', respectively.

Table 7.1: Example of life table for the codling moth in different apple and pear cultivars.

Initial number of migrating codling moth adult pairs = 10							
Gen.	Variable	Cultivar					
		'Red Delicious'		'Anjou'		'Bartlett'	
		%	No. viable individuals	%	No. viable individuals	%	No. viable individuals
1st	% Selecting host	35.0	3.5	30.0	3.0	35.0	3.5
	% Mating on host	100.0	3.5	85.0	2.6	90.0	3.2
	Oviposition level on host	127.0	444.5	118.4	301.9	116.9	368.2
	% Egg survivorship on host	92.4	410.7	91.9	277.5	92.4	340.2
	% Neonate larvae survivorship * (while searching for a fruit)	50.0	205.4	50.0	138.7	50.0	170.1
	% First-instar larvae survivorship (while attempting penetration)	61.1	125.5	22.3	30.9	57.4	97.7
	% Larval survivorship inside fruit	86.4	108.4	87.4	27.0	87.2	85.2
	% Pupal survivorship	100.0	108.4	100.0	27.0	100.0	85.2
2nd	% Mating on host	100.0	108.4	85.0	23.0	90.0	76.6
	Oviposition level on host	143.4	15546.0	131.9	3031.5	134.8	10330.7
	% Egg survivorship on host	89.9	13975.8	89.8	2722.3	90.6	9359.6
	% Neonate larvae survivorship * (while searching for a fruit)	50.0	6987.9	60.0	1633.4	60.0	5615.7
	% First-instar larvae survivorship (while attempting penetration)	68.3	4772.7	59.2	957.0	72.9	4093.9
	% Larval survivorship inside fruit	86.9	4147.5	87.8	849.0	84.7	3467.5
	% Overwintering larvae mortality **	41.0	1700.5	41.0	348.1	41.0	1421.7

* Assumed value (no estimate available)

** Estimate from MacLellan (1962), assuming no differences on number of overwintering sites among cultivars

The percentage of mating on host in Table 7.1 is the estimate of the proportion of adults that would successfully mate on trees of the cultivar selected as host. The estimates are 100, 85, and 90%, respectively on 'Red Delicious', 'Anjou', and 'Bartlett'. These values come from the mating experiments conducted in different host cultivars (see Fig. 4.3; Chapter 4).

Estimates of oviposition level on host in Table 7.1 are the values of the mean number of eggs/female obtained in the adult fecundity experiments presented in Table 6.3 (Chapter 6).

The estimates of the percentage of egg survivorship in Table 7.1 are the mean values of egg survival on the different oviposition surfaces (see Table 5.1; Chapter 5). Survival of eggs of the first generation was estimated from the experiments conducted on June. Survival of eggs of the second generation was estimated from the experiments conducted on July and August.

From the studies conducted in this research project no values of mortality of neonate larvae while searching for a fruit can be estimated. In the example presented in Table 7.1, assumptions were made that survivorship of neonate larvae in the process of searching for a fruit was 50% in all cultivars for individuals of the first generation, and 50% in 'Red Delicious' and 60% in 'Anjou' and 'Bartlett' pear for individuals of the second generation. A higher percentage of larvae reaching the fruit is assumed in pear cultivars late in the season because more eggs are laid directly on the fruit surface by moths of the second generation. Jackson (1982) studied the effect of temperature and relative humidity on first-instar larvae mortality. However, no data are available on the effect of distance from egg laying to fruit on neonate larvae mortality in different host cultivars.

The values of first-instar larval survivorship while attempting penetration and larval survival inside the fruit in Table 7.1 are estimated from the results of presented in Fig. 5.1 and Fig. 5.8 (Chapter 5), respectively. Estimates of first generation larvae successfully entering the fruit or emerging from the fruit are the mean survival values obtained in the larval mortality experiments conducted from May until early July. Estimates for the second generation are the mean survival values from the experiments conducted in August and September.

MacLellan (1962) estimated that the mortality of overwintering larvae was 59.0. Assuming that the characteristics of the trees of the tree cultivars are similar and they have the same number of overwintering sites, the same estimate can be used for the different cultivars.

The number of successful first-instar larvae penetrating the fruit gives an idea of the number of infested fruit in different host cultivars. In the example presented in Table 7.1, the predicted number of first-instar larvae successfully entering the fruit is significantly higher in 'Red Delicious' apple and 'Bartlett' pear (higher than 4,000) than in 'Anjou' pear (about 1,000). This agrees with the infestation levels observed in the field.

Oviposition on different host surfaces (fruit, upper side and underside of leaves, wood) has no direct effect on survival since there was differential egg mortality on different oviposition surfaces. However, egg distribution is indirectly correlated to neonate larval mortality since the probability of successfully locating a fruit can be expected to be higher on those cultivars the farther eggs are laid away from fruit and the farther larvae have to travel to reach a fruit. Therefore, egg distribution could be considered in the life table to assess neonate mortality while searching for fruit.

Other estimates that could be included in the life table are mortality of different stages due to natural enemies and insecticide intervention. Estimates of mortality due to predation and parasitism have been reported by several authors. MacLellan (1962) determined that egg predation in an apple orchard accounted for 14.4 % mortality and egg parasitism by *Trichogramma* sp. For 2.4 %. Mortality associated to insecticide sprays can be estimated knowing the product applied, time of application, and the distribution of insecticide and the pest in the tree.

A further step would be to use the life table estimates in sensitivity and key-factor analysis to investigate what factors contribute more in regulating codling moth populations and to test the consequences of manipulating variables that operate in different age intervals.

The research presented here has given a good idea of how host-specific characteristics affect several aspects of the biology and behavior of the codling moth that can explain differences in susceptibility among apple and pear cultivars. However, more information is needed for a better understanding of the relationship between the codling moth and its hosts and the factors responsible for resistance to codling moth attack.

The present study has shown that laboratory strains of the codling moth reared on artificial diet and on apples behave similarly. However, no release experiments were done with feral moths. Additional studies should be conducted with feral moths of different apple and pear strains to compare their behavior to the laboratory strains used in this study. Future research should investigate whether or not the formation of host races has any effect on the behavior of the moth during host selection. In addition, release experiments were conducted in a complete artificial situation (mixed trees of different species and host cultivars). However, this layout is not found in commercial orchards. Similar preference experiments can be conducted with moths exposed to blocks of different species and host cultivars to see whether or not the behavior pattern is similar to that observed in our experiments. It would also be of interest to investigate whether or not differences in susceptibility among hosts not closely related (pome fruit, such as apple and pear, versus stone fruit, such as apricot and plum, trees) are a manifestation of adult preference. In the research presented here, the role of different chemical volatiles release by host plants (including alpha-farnesene) has not been investigated in depth. This is another area that should be worked on in the future.

Another aspect of the behavior of the codling moth that deserves attention is the role of stimuli from host plants in the induction of calling by adult females. Differences in stimuli among host cultivars could lead to different calling success and, consequently, to differences in mating success and oviposition levels. The factors affecting egg laying and oviposition site selection have been widely studied and are quite known. However, there are some aspects still not understood. The role of chemical volatiles (alpha-farnesene, ethylene) on mating/oviposition also needs to be studied more in depth. Higher egg-laying on leaf surfaces facing a host fruit can be related to a higher content (production and/or absorption) of adult attractants and oviposition stimulants (such as alpha-farnesene) in the wax layer of leaf surfaces facing a fruit have. Studies should be conducted with feral and laboratory moths of different strains to compare their behavior.

There is contradictory information about the behavior of neonate larvae while searching for a fruit. Results showed that visual stimuli from host fruit are relatively more important than olfactory stimuli to orient a neonate larvae to a fruit. However, experiments were conducted with individuals of a laboratory strain reared on apple. The behavior of

field individuals may be different and should be investigated. In addition, there is little information on the efficiency and mortality of first-instar larvae while searching for a fruit on natural branches on different host cultivars. This phase in the life cycle is critical for codling moth survivorship when eggs are not laid on the fruit surface. The present study has given a good understanding of the resistance factors associated with codling moth resistance to larval penetration. However, more research is needed to try to determine some physical and/or chemical characteristics of the fruit that confer resistance to larval penetration through the side of the fruit, especially on pears. Electron-microscopy could be a very useful tool in this respect. The determination of the mortality of overwintering larvae in individuals reared on different host fruits is unknown and deserves more attention. Overwintering larvae mortality can also be a key factor to explain codling moth population dynamics and infestation levels in different host cultivars.

Results have shown that larval food has no effect on larval and pupal developmental rates. However, the experiments were conducted with individuals of a laboratory strain and no consecutive generations were reared on fruit of the pear cultivars tested. Host races can indeed have different heat requirements. Therefore, larval and pupal development of strains adapted to apple and pear cultivars should be studied to answer this question. If apple and pear races have different heat requirements, phenology models developed from data with apple as food source would not be valid for pear orchards.

BIBLIOGRAPHY

- Anet, E. F. L. J. 1969.** Autoxidation of alpha-farnesene. *Aust. J. Chem.* 22: 2403-2410.
- Anet, E. F. L. J. 1970.** Synthesis of (E,Z)-alpha, (Z,Z)-alpha, and (Z)-beta-farnesene. *Aust. J. Chem.* 23: 2101-2108.
- Armstrong, T. 1945.** Differences in the life history of codling moth, *Carpocapsa pomonella* (L.), attacking pear and apple. *Can. Entomol.* 77: 231-233.
- Audemard, H. 1976.** Étude demoécologique du carpocapse (*Laspeyresia pomonella* L.) en verger de pommiers de la basse vallée du Rhône et possibilités d'organisation d'une lutte intégrée. Doctoral dissertation, Univ. Tours., France. 145 pp.
- Bäckman, A. C. 1999.** Olfactory communication in the codling moth, *Cydia pomonella* L. Doctoral dissertation, Swedish University of Agricultural Sciences. 111 pp.
- Bäckman, A. C., M. Bengtsson, J. Löfqvist & P. Witzgall. 1998.** Plant volatile compounds from apple, pp. 2. *In* Proceedings, Use of pheromones and other semiochemicals in integrated control. Scents in orchards – Symposium on plant and insect semiochemicals from orchard environments, 21-24 September 1998. IOBC wprs Working Group Meeting. Dachau, Germany.
- Balensky, A., S. Ivanor & I. Ivanor. 1973.** *Laspeyresia pomonella* -an enemy also of almond. *Dokl. Sel'skokhozyais tvennoi Akad. im. Georgiya Dimitrova* 6(3): 201-204.
- Barnes, M. M. 1991.** Codling moth occurrence, host race formation, and damage, pp. 313-327. *In* L. P. S. Van der Geest & H. H. Evenhuis [eds.], *Tortricid pests, their biology, natural enemies and control*. Elsevier Science Publishers, Amsterdam, The Netherlands
- Batiste, W. C., W. H. Olson & A. Berlowitz. 1973.** Codling moth: influence of temperature and daylight intensity on periodicity of daily flight in the field. *J. Econ. Entomol.* 66(4): 883-892.
- Bauckmann, M. 1956.** Untersuchungen über eine termingerechte Bekämpfung des Apfelwicklers (*Carpocapsa pomonella* L.) unter Berücksichtigung des Falterfluges. *Arch. f. Gartenbau* 4: 235-276.
- Bernasconi, M. L., T. C. J. Turlings, L. Ambrosetti, P. Bassetti & S. Dorn. 1998.** Herbivore-induced emissions of maize volatiles repel the corn leaf aphid, *Rhopalosiphum maidis*. *Entomol. exp. et appl.* 87: 133-142.
- Beyer, E. M. & P. W. Morgan. 1970.** A method for determining the concentration of ethylene in the gas phase of vegetative plant tissues. *Plant Physiol.* 46: 352-354.

- Blomefield, T. L. 1991.** Codling moth management: there's more to it than spraying. *Decid. Fr. Grow.* 41: 250-251.
- Blomefield, T. L. 1994.** Codling moth resistance: is it here, and how do we manage it? *Decid. Fr. Grow.* 44: 130-132.
- Blomefield, T. L. 1995.** Mating disruption of codling moth. *Proc. Ann. Mtg. Cape Pomological Assoc.*: 50-55.
- Blomefield, T. L., K. L. Pringle & A. Sadie. 1997.** Field observations on oviposition of codling moth, *Cydia pomonella* (Linnaeus) (Lepidoptera: Olethreutidae), in an unsprayed apple orchard. *Afr. Entomol.* 5: 319-336.
- Boevé, J. L., U. Lengwiler, L. Tollsten, S. Dorn & T. C. J. Turlings. 1996.** Volatiles emitted by apple fruitlets infested by larvae of the European apple sawfly. *Phytochemistry* 42(2): 373-381.
- Borden, A. D. 1931.** Some field observations on codling moth behavior. *J. Econ. Entomol.* 24(6): 1137-1145.
- Bovey, P. 1949.** Le carpocapse des pommes (*Enarmonia pomonella* L.) ravageur des abricots en Valais. *Suisse Stat. Féd. D'essais vit., abor. Et de chim. Agric. Publ.* 381: 137-172.
- Bradley, S. J. & D. M. Suckling. 1995.** Factors influencing codling moth larvae response to alpha-farnesene. *Entomol. exp. et appl.* 75(3): 221-227.
- Brunner, J. F. & S. C. Hoyt. 1982.** Codling moth control- A new tool for timing sprays. *Washington State University Cooperative Extension Bulletin* 1072. 4 pp.
- Buda, V. 1978.** Host plant odour in codling moths' pheromone communication. In "Tezisi II konferentsii molodykh utshenykh". Vilnius (in Russian).
- Castrovillo, P. J. & R. T. Cardé. 1979.** Environmental regulation of female calling and male pheromone response periodicities in the codling moth (*Laspeyresia pomonella*). *J. Insect Physiol.* 25: 659-667.
- Castrovillo, P. J. & R. T. Cardé. 1980.** Male codling moth (*Laspeyresia pomonella*) orientation to visual cues in the presence of pheromone and sequences of courtship behaviors. *Ann. Entomol. Soc. Am.* 73: 100-105.
- Chapman, P. J. & S. E. Lienk. 1971.** Tortricid fauna of apple in New York (Lepidoptera: Tortricidae); including an account of apples occurrence in the state especially as a naturalized plant. *A Special Publication, N.Y.State Agr. Expt. Sta., Geneva, N.Y.*

- Chernyshev, V. B. 1961.** Vremya leta razlichnykh nasekomykh na svet (Time of flight to light of various insects). Zool. Zhurn. 40(7): 1008-1019.
- Cisneros, F. H. & M. M. Barnes. 1974.** Contribution to the biological and ecological characterization of apple and walnut host races of codling moth, *Laspeyresia pomonella* (L.): moth longevity and oviposition capacity. Environ. Entomol. 3: 402-406.
- Coutin, R. 1959.** La penetration des larves de *Laspeyresia pomonella* L. dans les fruits des Pomacees. Bull. Soc. Entomol. Fr. 64: 100-105.
- Croft, B. A. & H. W. Riedl. 1991.** Chemical control of the codling moth, pp. 371-388. In L. P. S. van der Geest & H. H. Evenhuis [eds.], World crop pests. Tortricid pests: their biology, natural enemies and control. Elsevier, Amsterdam, Holland.
- Crosby, J. A., J. Janick, P. Pecknold, S. S. Korban, P. A. O'Connor, S. M. Ries, J. Goffreda & A. Voode. 1997.** Breeding apples for scab resistance: 1945-1990. Fruit varieties journal 46: 145-166.
- Curtis, C. E., J. S. Tebbets & J. D. Clark. 1990.** Ovipositional behavior of the codling moth (Lepidoptera: Tortricidae) on stone fruits in the field and an improved oviposition cage for use in the laboratory. J. Econ. Entomol. 83(1): 131-134.
- Cutright, C. R. 1931.** Some laboratory reactions of young codling moth larvae. J. Econ. Entomol. 24: 81-83.
- Cutright, C. R. 1937.** Codling moth biology and control. Ohio Agr. Sta. Bull. 583.
- Cutright, C. R. 1954.** A codling moth population resistance to DDT. J. Econ. Entomol. 47(1): 189-190.
- Cutright, C. R. & H. E. Morrison. 1935.** Varietal susceptibility to codling moth injury. J. Econ. Entomol. 28: 107-109.
- De Jong, D. J. & J. J. Post. 1963.** Influence of weather on the development and activity of the Lepidoptera concerned, pp. 41-84. In H.J. De Fluiter, P.H. Van de Pol and J.P.M. Woudenberg [Eds.], Phenological and faunistic investigations on orchard insects. Versl. Landbouwk. Onderz., Wageningen, Netherl. 69(14), 226 pp.
- Deseő, K. V. 1970.** The effect of olfactory stimuli on the oviposition behaviour and egg production of some micro-lepidopterous species. Colloques int. CEntomol. Natn. Rech. SciEntomol. 189: 163-174.
- Deseő, K. V. 1971.** Study of factors influencing the fecundity and fertility of codling moth ((*Laspeyresia pomonella* L.), Lepid.; Tortr.). Acta Phytopathologica Academiae Scientiarum Hungaricae 6: 243-252.

- Deseő, K. V. 1973.** Reproductive activity of codling moth (*Laspeyresia pomonella* L., Lepidopt.; Tortr.) exposed to short photophase during preimaginal state. *Acta Phytopathologica Academiae Scientiarum Hungaricae* 8(1-2): 193-206.
- Deseő, K. V. & G. Y. Saringer. 1975a.** Photoperiodic effect on fecundity of *Laspeyresia pomonella*, *Grapholita funebrana* and *G. molesta*: the sensitive period. *Entomol. exp. et appl.* 18: 187-193.
- Deseő, K. V. & G. Y. Saringer. 1975b.** Photoperiodic regulation in the population dynamics of certain lepidopterous species. *Acta Phytopathologica Academiae Scientiarum Hungaricae* 10(1-2): 131-139.
- Dethier, V. G. 1947.** Chemical Insect Attractants and Repellents. The Blakiston Company, Philadelphia, Pa. 289 pp.
- Dickson, R. C., M. M. Barnes & C. L. Turcan. 1952.** Continuous rearing of the codling moth. *J. Econ. Entomol.* 45: 66-68.
- Ebel, R. C., J. P. Mattheis & D. A. Buchanan. 1995.** Drought stress of apple trees alters leaf emissions of volatile compounds. *Physiol. Plant.* 93: 709-712.
- Falcon, L. A. & J. Huber. 1991.** Biological control of the codling moth. pp. 339-370. *In* L. P. S. van der Geest & H. H. Evenhuis [eds.], *World crop pests. Tortricid pests: their biology, natural enemies and control*. Elsevier, Amsterdam, Holland.
- Felt, E. P. 1910.** Recent experiments with codling moths. *J. Econ. Entomol.* 3: 474-477.
- Ferro, D. N. & Akre, R. D. 1975.** Reproductive morphology and mechanics of mating of codling moth, *Laspeyresia pomonella*. *Ann. Entomol. Soc. Am.* 68: 417-424.
- Ferro, D. N., R. R. Sluss & T. P. Bogyo. 1975.** Factors contributing to the biotic potential of the codling moth, *Laspeyresia pomonella* (L.), in Washington. *Environ. Entomol.* 4: 385-391.
- Frick, K. E., H. G. Simkover & H. S. Telford. 1954.** Bionomics of the cherry fruit flies in eastern Washington. *Wash. Agric. Exp. Stn. Tech. Bull.* 13.
- Garlick, W. G. 1940.** The migration of codling moth larvae from one apple to another. *Can. Entomol.* 72: 87.
- Garlick, W. G. 1948.** A five-year field study of codling moth habits and adult emergence. *Scientific Agriculture* 28(7): 273-292.
- Garlick, W. G. & H. R. Boyce. 1940.** A note on the habits of mature codling moth larvae. *Can. Entomol.* 72: 87.

Gehring, R. D. & H. S. Madsen. 1963. Some aspects of mating and oviposition behavior of the codling moth, *Carpocapsa pomonella*. J. Econ. Entomol. 56: 140-143.

Geier, P. W. 1963. The life history of codling moth *Cydia pomonella* (L.) (Lepidoptera: Tortricidae) in the Australian Capital Territory. Aust. J. Zool. 11: 323-367.

Gentner, L. G. 1940. Spur-borrowing habit of the codling moth larvae on pear trees. J. Econ. Entomol. 33(5): 796-799.

Glenn, P.A. 1922. Relation of temperature to development of the codling moth. J. Econ. Entomol. 15: 193-199.

Goonewardene, H. F., W. F. Kwolek & R. A. Hayden. 1984. Survival of immature stages of the codling moth (Lepidoptera: Tortricidae) on seeded and seedles apple fruit. J. Econ. Entomol. 77(6): 1427-1431.

Gut, L. J. 1980. Oviposition of the codling moth, *Laspeyresia pomonella* (L.) on several varieties of apples and pears. Master dissertation. Oregon State University, Corvallis. 124 pp.

Hagley, E. A. C. 1969. The distribution and survival of overwintering codling moth larvae in southern Ontario. Proceedings of the Entomological Society of Ontario 100: 40-47.

Hagley, E. A. C. 1972a. Observations on codling moth longevity and egg hatchability. Environ. Entomol. 1: 123-125.

Hagley, E. A. C. 1972b. Effect of rainfall on the survival and establishment of codling moth larvae. Environ. Entomol. 4: 446-447.

Hagley, E. A. C. 1976. Effect of rainfall and temperature on codling moth oviposition. Environ. Entomol. 5(5): 967-969.

Hagley, E. A. C., J. F. Bronskill & E. J. Ford. 1980. Effect of the physical nature of leaf and fruit surfaces on oviposition by the codling moth, *Cydia pomonella* (Lepidoptera: Tortricidae). Can. Entomol. 112(5): 503-510.

Hall, J. A. 1929. Six years's study of the life history and habits of the codling moth (*Carpocapsa pomonella* (L.)). Annual report of the Entomological Society of Ontario 59: 96-105.

Hall, J. A. 1934. Observations on the behaviour of newly hatched codling moth larvae. Can. Entomol. 66: 100-102.

Hamstead, E. O. & E. Gould. 1960. Codling moth oocyte studies. J. Econ. Entomol. 43: 724-726.

- Hathaway, D. O., A. E. Clift & B. A. Butt. 1971.** Development and fecundity of codling moths reared on artificial diets or immature apples. *J. Econ. Entomol.* 64(5): 1088-1090.
- Hattingh, C. C. 1942.** A study of codling moth oviposition in a mixed pear orchard. *J. Entomol. Soc. South Afr.* 6: 137-146.
- Hattingh, C. C. 1943.** The distribution of codling moth eggs on pear trees. *J. Entomol. Soc. South Afr.* 6: 124-130.
- Heriot, A. D. & D. B. Waddell. 1942.** Some effects of nutrition on the development of the codling moth. *Sci. Agr.* 23: 172-175.
- Hintze, J. L. 1997.** Number Cruncher Statistical System, version 97. Kaysville, UT.
- Hoerner, J. L. 1925.** Notes on codling moth larvae. *J. Econ. Entomol.* 18: 423-424.
- Hough, W. S. 1928.** Relative resistance to arsenical poisoning of two codling moth strains. *J. Econ. Entomol.* 21: 325-329.
- Howell, J. F. 1981.** Codling moth, *Cydia pomonella*: the effect of adult diet on longevity, fecundity, fertility, and mating. Various carbohydrates and proteins. *J. Econ. Entomol.* 74(1): 13-18.
- Howell, J. F. & A. D. Clift. 1974.** The dispersal of sterilized codling moth released in the Wenas Valley, Washington. *Env. Entomol.* 3: 75-81.
- Howell, J. F., R. B. Hutt & W. B. Hill. 1978.** Codling moth: mating behaviour in the laboratory. *Ann. Entomol. Soc. Am.* 71: 891-895.
- Huard, V. A. 1916.** Les principales especes d'insectes nuisibles et de maladies vegetales. Quebec. *Sci. Agric.* 20: 624-631.
- Huelin, F. E. & K. E. Murray. 1966.** Alpha-farnesene in the natural coating of apples. *Nature* 210: 1260-1261.
- Hutt, R. B. & L. D. White. 1977.** Mating response to visual stimulus in the male codling moth (*Laspeyresia pomonella*). *Environ. Entomol.* 6(4): 567-568.
- Isley, D. 1938.** Codling moth oviposition and temperature. *J. Econ. Entomol.* 31: 356-359.
- Isley, D. 1939.** Timing seasonal occurrence and abundance of the codling moth. *Arkansas Agr. Expt. Sta. Bull.* 382, pp. 1-26.
- Isley, D. & A. J. Ackerman. 1923.** Life history of the codling moth in Arkansas. *Univ. Ark. Agr. Sta. Bull.* 189.

- Jackson, D. M. 1976.** Behavior of first instars of the codling moth, *Carpocapsa pomonella*, pest of apples, pears, quince, and other tree fruit. Bull Oreg Entomol Soc. 61: 493-496.
- Jackson, D. M. 1979.** Codling moth egg distribution on unmanaged apple trees. Ann. Entomol. Soc. Amer. 72: 361-368.
- Jackson, D. M. 1982.** Searching behavior and survival of 1st-instar codling moths. Ann. Entomol. Soc. Amer. 75: 284-289.
- Jackson, D. M. & R. F. Harwood. 1980.** Survival potential of first instars of the codling moth in laboratory experiments. Ann. Entomol. Soc. Amer. 73: 160-163.
- Jennings, W. C. & R. Tressl. 1974.** Production of volatile compounds in the ripening Bartlett pear. Cehm Mikrobiol. Technol. Lebensm. 3: 52-55.
- Kennedy, J. S. 1977.** Olfactory responses to distant plants of attractants and repellents, pp. 215-229. In Shorey, H. H. & J. J. McKelvey, Jr. (Eds.). Chemical control of insect behavior. Theory and practice. Wiley Interscience. New York.
- Klinger, J., W. Vogel & H. Wille. 1958.** Der Einfluss der Temperatur auf die Eiablage des Apfelwicklers. Schweiz. Zeitsch. Obst - und Weinbau 67: 256-62.
- Knight, A. L. 1998.** Management of codling moth (Lepidoptera: Tortricidae) in apple with overhead watering. J. Econ. Entomol. 91(1): 209-216.
- Košťál, V. & S. Finch. 1994.** Influence of background on host-plant selection and subsequent oviposition by the cabbage root fly (*Delia radicum*). Entomol. exp. et appl. 70(2): 153-163.
- Košťál, V. & S. Finch. 1996.** Preference of the cabbage root fly, *Delia radicum* (L.) for coloured traps: influence of sex and physiological status of the flies, trap background and experimental design. Physiol. Entomol. 21(3): 123-130.
- Labeyrie V. 1960.** Influence de l'hôte sur la fécondité d'*Acanthoscelides obtectus* Say (Coleopteres - Bruchidae). C.R. Acad. Sci. 250: 615-617.
- Lacey, L. A. & R. L. Chauvin. 1999.** Entomopathogenic nematodes for control of diapausing codling moth (Lepidoptera: Tortricidae) in fruit bins. J. Econ. Entomol. 92(1): 104-109.

Landolt, P. J. 1998. Plant odorants in phytophagous insect host finding and host selection, pp. 10. *In* Proceedings, Use of pheromones and other semiochemicals in integrated control. Scents in orchards – Symposium on plant and insect semiochemicals from orchard environments, 21-24 September 1998. IOBC wprs Working Group Meeting. Dachau, Germany.

Landolt, P. J., R. W. Hofstetter & P. S. Chapman. 1998. Neonate codling moth larvae (Lepidoptera: Tortricidae) orient anemotactically to odor of immature apple fruit. *The Pan-Pacific entomologist* 74(3): 140-149.

Lekic, M. B. 1950. The biology of the codling moth in the territory of the Serbian People's Republic and measures of its control. *Zast. Bilja* 1(1): 32-65 [Rev. Appl. Entomol. Ser. A. 41: 104].

Lekic, M. B. 1958. O Nekim pitanjima in biologije jabukinog smotavca. *Eastita Bilja* 45: 89-184.

Logan, J. A., D. J. Wollkind, S. C. Hoyt & L. K. Tanigoshi. 1976. An analytic model for description of temperature dependent rate phenomena in arthropods. *Environ. Entomol.* 5: 1133-1140.

MacLellan, C. R. 1960. Cocooning behaviour of overwintering codling moth larvae. *Can. Entomol.* 92: 469-479.

MacLellan, C. R. 1962. Mortality of codling moth eggs and young larvae in an integrated control orchard. *Can. Entomol.* 94: 655-666.

Madsen H. F. & A. D. Borden. 1954. Codling moth and orange tortrix control on apricots in California, 1948-1953. *J. Econ. Entomol.*: 161-165.

Mailloux, M. & E. J. LeRoux. 1960. Further observations on the life-history and habits of the codling moth, *Carpocapsa pomonella* (L.) (Lepidoptera: Tortricidae), in apple orchards in south-western Quebec. *Annu. Rep. Quebec Pomol. Fruit Grow. Soc.* 60: 45-56.

Mani, E. & T. Wildbolz. 1977. The dispersal of male codling moths (*Laspeyresia pomonella* L.) in the Upper Rhine Valley. *Z. angew. Ent.* 83: 161-168.

Mantey, D. & L. D. White (Short Communications). Removal of intact female reproductive organs from codling moths.

Marshall, G. E. 1940. Some newly discovered habits of the codling moth. *J. Econ. Entomol.* 33(1): 200.

Martí, S. 1995. Desenvolupament i diapausa de *Cydia pomonella* (L.) (Lepidoptera: Tortricidae) en condicions de camp a la zona de Lleida. PFC dissertation. Universitat de Lleida, Spain. 147 pp. (In Catalan).

- Martí, S., D. Bosch, M. J. Sarasúa & J. Avilla. 1996.** Validation of linear and non-linear models of the rate of development of a Spanish population of *Cydia pomonella*. IOBC/WPRS Bulletin 19(4): 118-122.
- McIndoo, N. E. 1928.** Tropic responses of codling moth larvae. J. Econ. Entomol. 21(4): 631.
- McIndoo, N. E. 1929.** Tropisms and sense organs of Lepidoptera. Smithson. Misc. Collect. 81: 1-59.
- Meigh, D. F. & A. A. E. Filmer. 1967.** Chemical investigation of superficial scald in stored apples. Rep. Agric. Res. Cin. (Ditton Laboratory) (1966-1967). pp 36-37.
- Meir, S. & W. J. Bramlage. 1988.** Antioxidant activity in "Cortland" apple peel and susceptibility to superficial scald after storage. J. Amer. Soc. Hort. Sci. 113(3): 412-418.
- Mellenthin, W. M. & D. Bonney. 1972.** A portable limb enclosure for temperature modification of tree fruits. HortScience 7(2): 134-136.
- Murray, K. E., F. E. Huelin & J. B. Davenport. 1964.** Occurrence of farnesene in the natural coating of apples. Nature 204: 80.
- Newcomer, E. J. & M. A. Yothers. 1924.** Control of the codling moth in the Pacific Northwest. Farmers Bull. U.S. Dept. Agri. 1326.
- Nikolic, D. 1958.** Klimatiste prilike i pojava jabukinog smotavca u Tetovu u toku petogodisneg perioda (1952-1956). Zastita Bilja 45: 67-74.
- Nowosielski, J. W. & Z. W. Suski. 1977.** Observations on the mating behaviour of the codling moth, *Laspeyresia pomonella* (L.). II. Temporal patterns of copulatory behaviour in relation to the age of the moths and the time of day. Ekologia Polska 25(2): 341-352.
- Oatman, E. R. 1964.** Apple maggot trap and attractant studies. J. Econ. Entomol. 57: 529-531.
- Olson, W. H. 1977.** Walnut varieties differ in susceptibility to codling moth damage. Calif. Agric. 31: 14-15.
- Onstad, D. W. & W. H. Reissig. 1986.** Influence of apple cultivar, tree phenology, and leaf quality on the development and mortality of *Choristoneura rosaceana* (Lepidoptera: Tortricidae). Can. Entomol. 78: 1455-1462.
- Park, K. T. & J. S. Park. 1976.** The absence of codling moth, *Cydia pomonella* L., in Korea. Korean Journal of Plant Protection 15: 79-81.

- Parker, R. L. 1959.** Notes on oviposition behavior responses of the codling moth, *Carpocapsa pomonella* L., to air movement, temperature, and light. J. Kan. Entomol. soc. 32: 152-154.
- Parrot, P. J. & D. L. Collins. 1934.** Phototropic responses of the codling moth. J. Econ. Entomol. 27: 370-79.
- Petty, F. W. 1926.** Codling moth in apricots. Preliminary report on the biology of the codling moth and its control in apricots, Wellington, during the 1924-25 season. J. Dept. Agr. S. Africa. 11: 56-65, 137-152.
- Phillips, P. A. & M. M. Barnes. 1975.** Host race formation among sympatric apple, walnut, and plum populations of the codling moth, *Laspeyresia pomonella* (L.). Ann. Entomol. Soc. Am. 68(6): 1053-1060.
- Pickel, C., R. S. Bethell & W. W. Coates. 1986.** Codling moth management using degree days. Univ. Calif. Integr. Pest Manage. Publ. 4.
- Pitcairn, M. J., C. Pickel, L. A. Falcon & F. G. Zalom. 1991.** Development and survivorship of *Cydia pomonella* (L.) (Lepidoptera: Tortricidae) at ten constant temperatures. The Pan-Pacific Entomologist 67(3): 189-194.
- Plourde, D. F., H. F. Goonewardene & W. F. Kwolek. 1985.** Pubescence as a factor in codling moth oviposition and fruit entry in five apple selections. Hortsci. 20: 82-84.
- Pristavko, V. P. 1969.** Evaluation of some abiotic factors influencing captures of coding moths by black-light traps. J. Zool. 48: 1177-84.
- Pristavko, V. P. 1971.** Daily flight activity and migration range of the codling moth (*Laspeyresia pomonella*) in the steppe and forest-steppe zones of Ukraine. Zool. Z.H. 50(1): 67-71.
- Pristavko, V. P. & V. M. Chaika. 1976.** Olfactory sensitivity of the codling moth as related to insect age, air temperature, and time of day: Electrophysiological evaluation. Dopov. Akad. Nauk. UKR, RSR (SER B) Heol Khim Biol. Nauky 7: 650-52.
- Prokopy, R. J. 1968.** Visual responses of apple maggot flies, *Rhagoletis pomonella* (Diptera: Tephritidae): orchard studies. Entomol. exp. et appl. 11: 403-422.
- Prokopy, R. J. 1972.** Response of apple maggot flies to rectangles of different colors and shades. Environ. Entomol. 1: 720-726.
- Prokopy, R. J., R. J. Collier & S. Finch. 1983.** Visual detection of host plants by cabbage root flies (*Delia radicum*). Entomol. exp. et appl. 34(1): 85-89.

Provancher, L. L. 1874. Le verger, le potager et le parterre dans la province de Quebec. C. Darveau (Eds.), Quebec. 332 pp.

Proverts, M. D. & J. R. Newton. 1962. Effect of heat on the fertility of the codling moth, *Carpocapsa pomonella* (L.) (Lepidoptera: Olethreutidae). Can. Entomol. 94: 225-233.

Putman, W. L. 1963. The codling moth *Carpocapsa pomonella* (L.) (Lepidoptera: Tortricidae): A review with special reference to Ontario. Proc. Entomol. Soc. Ont. 93: 22-60.

Quayle, H. J. 1926. The codling moth in walnuts. Univ. Calif. Agr. Exp. Sta. Bull. 402.

Riedl, H. 1983. Analysis of codling moth phenology in relation to latitude, climate and food availability, pp. 233-252. In V. K. Brown & I. Hodek [eds.], Diapause and life cycle strategies in insects. Dr. W. Junk Publishers, The Hague, Belgium.

Riedl, H. & B. A. Croft. 1975. Use of the pheromone trap to quantitatively assess the phenology and density of the codling moth. Proc. National Pome and Stone Fruit Extension Pest Management Workshop, Yakima, Wash., March 11-12, 1975, pp.58-67.

Riedl, H. & B. A. Croft. 1978. The effects of photoperiod and effective temperatures on the seasonal phenology of the codling moth (Lepidoptera: Tortricidae). Can. Entomol. 110: 455-570.

Riedl, H. & W. Loher. 1980. Circadian control of oviposition in the codling moth, *Laspeyresia pomonella*, Lepidoptera: Olethreutidae. Entomol. exp. et appl. 27: 38-49.

Riedl, H. & R. Hislop. 1985. Visual attraction of the walnut husk fly (Diptera: Tephritidae) to color rectangles and spheres. Environ. Entomol. 14: 810-814.

Riedl, H. & R. Zelger. 1994. Erste Ergebnisse der Untersuchungen zur Resistenz des Apfelwicklers gegenüber Diblubenuron. Obstbau Weinbau 31: 107-109.

Riedl, H., S. A. Hoying, W. W. Barnett & J. E. DeTar. 1979. Relationship of within-tree placement of the pheromone trap to codling moth catches. Environ. Entomol. 8: 765-769.

Riedl, H., J. F. Howell, P. S. McNally & P. H. Westigard. 1986. Codling moth management: Use and standardization of pheromone trapping systems. Western Regional Research Publication, University of California. 23 pp.

Riedl, H., T. L. Blomefield & J. H. Giliomee. 1998. A century of codling moth control in South Africa: II. Current and future status of codling moth management. Entomol. J. S. Afr. Soc. Hort. Sci. 8(2): 32-54.

- Robert, P. 1964.** Influence de la plante-hôte sur l'activité de ponte de la teigne de la betterave, *Scrobipalpa* (Phtorimaea) ocellatella Boyd. *Ann. Epiphyties* 15: 325-326.
- Rock, G. C. & P. L. Shaffer. 1983.** Development rates of codling moth, *Cydia pomonella*, (Lepidoptera: Olethreutidae) reared on apple at four constant temperatures. *Environ. Entomol.* 12(3): 831-834.
- Russ, K. 1961.** Einfluss wichtiger Witterungsfaktoren auf die Flugtätigkeit des Apfelwicklers *Carpocapsa pomonella* L. *Pflanzenschutzberichte* 27: 67-82.
- Russ, K. 1976.** Investigations on the influence of fruit odour on the orientation of codling moth (*Laspeyresia pomonella*). *Symp. Biol. Hung.* 16: 237-240.
- Sanders, W. 1962.** Das Verhalten der Mittelmeerfruchtfliege *Ceratitis capitata* Wied. bei der Eiablage. *Z. Tierphysiol.* 19: 1-28.
- Saringer, Gy. 1963.** Autokologiai és ingerelettyini kutatások a maktokormanyossal (*Ceutorrhynchus macula-alba* Hrbst) kapcsoltatban. XIII. Congr. of Plant. Prot. Budapest.
- SAS Institute. 1990.** SAS/STAT User's Guide, 6th ed. SAS Institute, Cary, NC.
- SAS Institute. 1997.** SAS/STAT Software: Changes and enhancements through release 6.12. SAS Institute, Cary, NC.
- Sauphanor, B., M. Benoit, J. C. Bouvier, G. Perron, S. Malezieux & J. C. Fremond. 1994.** Un cas de résistance des pommes au diflubenzuron dans le sud-est de la France. *Phytoma-La-Defense-des-Vegeteux* (France) 458: 46-49.
- Sauphanor, B., J. C. Bouvier, V. Brose, K. Marty & S. Klier. 1996.** Characterization et detection de la résistance aux insecticides chimiques chez la carpocapse. *Phytoma* 482: 16-18.
- Sauphanor, B., C. Monier, S. Clier, J. C. Bovier & C. Martinet. 1997.** Extension en France de la résistance du carpocapse des pommes aux insecticides. *Annales ANPP. Quatrième conférence internationale sur les ravageurs en agriculture*, 1: 77-83.
- Schoonhoven, L. M. 1968.** Chemosensory bases of host plant selection. *A. Rev. Entomol.* 13: 115-136.
- Seaber, D., J. Strand & A. J. Strawn. 1990.** Degree-Day Utility (DDU) User-s Guide. UC IPM Publication 9, Division of Agriculture and Natural Resources. University of California. 127 pp.
- Serra, V. J. 1994.** Desarrollo e induccion de diapausa en *Cydia pomonella* (L.) (Lepidoptera: Tortricidae). PFC dissertation. Universitat de Lleida, Spain. 62 pp.

- Setyobudi, L. 1989.** Seasonality of codling moth, *Cydia pomonella* L. (Lepidoptera: Olethreutidae) in the Willamette Valley of Oregon: Role of photoperiod and temperature. Ph. D. dissertation, Oregon State University, Corvallis. 129 pp.
- Shel'Deshova, G. G. 1967.** Ecological factors determining distribution of the codling moth, *Laspeyresia pomonella* L. (Lepidoptera: Tortricidae) in the northern and southern hemispheres. Entomol. Rev. 46: 349-361.
- Shelford, V. E. 1927.** An experimental investigation of the relationships of the codling moth to weather climate. III. Nat. Hist. Surv. Bull. 16: 311-440.
- Shimizu, S. & S. Yoshihara. 1977.** The constituents of the essential oil from Japanese quince fruit, *Cydonia oblonga* Miller. Agric. Biol. Chem. 41(8): 1525-1527.
- Siegler, E.H. 1940.** Laboratory studies of codling moth larval attractants. J. Econ. Entomol. 33: 342-345.
- Siegler, E. H. & Plank, H. K. 1921.** Life history of the coding moth in the Grand Valley of Colorado. USDA Bull 932: 119 pp.
- Skirkevicius, A. & L. Tatjanskaite. 1971.** The sensitivity of the moth *Carpocapsa pomonella* L. to geraniol in connection with the different period of day. Proc. 1st All-Union Symp. On Insect Chemoreception, Vilnius, U.S.R.R.: 133-138.
- Skirkevicius, A. V. , V. Buda, G. H. L. Rothschild & Z. Skirkeviciene. 1980.** The response of male codling moths (*Laspeyresia pomonella*) to naturally emitted host plant odours. Short communications. Entomol. exp. et appl. 28: 334-338.
- Smith, C. M. 1989.** Plant resistance to insects. A fundamental approach. A Wiley-Interscience Publication. John Wiley and Sons. 286 pp.
- Song, Y. H. & H. Riedl. 1985.** Effects of temperature and photoperiod on male activity in *Laspeyresia pomonella* (L.) in New York. Korean J. Plant Prot. 24: 71-77.
- Speyer, W. 1932.** Can the codling moth develop on leaves alone? Arb. Biol. Reichsanst. Land u. Forstw. 20(2): 183-191.
- Steiner, L. F. 1939.** Distances traveled by newly hatched codling moth larvae. J. Econ. Entomol. 32(3): 470.
- Still, G. W. 1960.** An improved trap for deciduous tree fruit flies. J. Econ. Entomol. 53: 967.
- Subinprasert, S. & Svensson, B. W. 1988.** Effects of predation on clutch size and egg dispersion in the codling moth *Laspeyresia pomonella*. Ecol. Entomol. 13: 87-94.

Summerland, S. A. & L. F. Steiner. 1943. Codling moth oviposition and fate of the eggs. *J. Econ. Entomol.* 36(1): 72-75.

Suomi, D., J. J. Brown & R. D. Akre. 1986. Responses to plant extracts of neonatal codling moth larvae, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae: Olethreutinae). *J. Entomol. Soc. British Columbia* 83: 12-18.

Suski, Z. W., G. S. Labanowski & Z. Labanowski. 1981. Dispersal of laboratory-reared codling moth males, *Laspeyresia pomonella* (L.), in central Poland Ecology and management of pests in apple orchards. *Ekologia polska* (=Polish J. Ecol) 29(3): 441-449.

Sutherland, O. R. W. 1972. The attraction of newly hatched codling moth (*Laspeyresia pomonella*) larvae to apple. *Entomol. exp. et appl.* 15: 481-487.

Sutherland, O. R. W. & R. F. N. Hutchins. 1972. Alpha-farnesene, a natural attractant for codling moth larvae. *Nature* 239: 170.

Sutherland, O. R. W. & R. F. N. Hutchins. 1973. Attraction of newly hatched codling moth larvae (*Laspeyresia pomonella*) to synthetic stereoisomers of farnesene. *J. Insect Physiol.* 19: 723-727.

Sutherland, O. R. W., R. F. N. Hutchins & C. H. Wearing. 1974. The role of the hydrocarbon alpha-farnesene in the behavior of codling moth larvae and adults, pp. 249-263. *In* Browne, L.B. (Eds.) *Experimental analysis of insect behavior*. Springer Verlag. New York.

Sutherland, O. R. W., C. H. Wearing & R. F. N. Hutchins. 1977. Production of alpha farnesene, an attractant and oviposition stimulant for codling moth, by developing fruit of ten varieties of apple. *J. Chem. Ecol.* 3: 625-632.

Thiéry, D., B. Gabel, P. Farkas & M. Jarry. 1995. Egg dispersion in codling moth: influence of egg extract and of its fatty acid constituents. *J. Chem. Ecol.* 21(12): 2015-2026.

Trottier, R. & E. A. C. Hagley. 1979. Influence of temperature and snowfall on codling moth [*Laspeyresia pomonella*] fecundity in apple orchards (Ontario). *Environ. Entomol.* 8(6): 1052-1054.

Turlings, T. C. J., M. Bernasconi, R. Bertossa, F. Bigler, G. Caloz & S. Dorn. 1998. The induction of volatile emissions in maize by three herbivore species with different feeding habits: possible consequences for their natural enemies. *Biological control* 11: 122-129.

Van Leeuwen, E. R. 1939. Life history of the codling moth in northern Georgia. *USDA Tech. Bull.* 90: 1-94.

- Van Leewen, R. R. 1947.** Increasing production of codling moth eggs in an oviposition chamber. *J. Econ. Entomol.* 40: 744-745.
- Vermeulen, J., L. Cichón & E. Parra. 1989.** Sistema de alarma termoacumulativo para el control de carpocapsa (*Cydia pomonella*) para el Alto Valle del Río Negro y Neuquen. Instituto Nacional de Tecnología Agropecuaria. Estación Experimental Agropecuaria Alto Valle. Argentina. 14 pp.
- Vickers, R. A. 1997.** Effect of delayed mating on oviposition pattern, fecundity and fertility in codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae). *Australian J. Entomol.* 36: 179-182.
- Ville, C. A. 1967.** Biology. Mei Ya Publications, Inc. Taipei, Taiwan.
- Wearing, C. H. 1975a.** Integrated control of apple pests in New Zealand. 3. Natural mortality of fifth-instar larvae of codling moth tagged with cobalt-58 in relation to their distribution. *N. Z. J. Zool.* 2(1): 151-168.
- Wearing, C. H. 1975b.** Integrated control of apple pests in New Zealand. 2. Field evaluation of fifth-instar larval and pupal mortalities of codling moth by tagging with cobalt-58. *N. Z. J. Zool.* 2(1): 135-149.
- Wearing, C. H. & A. M. Ferguson. 1971.** Variation in the fecundity of the codling moth, *Laspeyresia pomonella* L. *N. Z. J. Sci.* 14(2): 233-237.
- Wearing, C. H. & R. F. N. Hutchins. 1973.** Alpha-farnesene, a naturally occurring oviposition stimulant for the codling moth, *Laspeyresia pomonella*. *J. Insect Physiol.* 19: 1251-1256.
- Wearing, C. H., P. J. Connor & K. D. Ambler. 1973.** Olfactory stimulation of oviposition and flight activity of the codling moth *Laspeyresia pomonella*, using apples in an automated olfactometer. *N. Z. J. Sci.* 16: 697-710.
- Weissling, T. J. & A. L. Knight. 1994.** A passive trap for monitoring codling moth (Lepidoptera: Tortricidae) flight activity. *J. Econ. Entomol.* 87(1): 103-107.
- Welter, S. C., J. E. Dunley, K. Hansen & M. Smirle. 1992.** Pesticide resistance in codling moth: cross-resistance, resistance in field populations, and genetic selection. 1992 Research Reports, California Pear Advisory Board, Sacramento, Calif.
- Westigard, P. H, L. G. Gentner & B. A. Butt. 1976.** Codling moth: egg and first instar mortality on pear with special reference to varietal susceptibility. *Environ. Entomol.* 5: 51-54.
- White, A. G. & V. G. Buss. 1998.** Breeding commercial apple cultivars in New Zealand with resistances to pests and diseases. *Acta horticulturae* 484: 157-161.

White, L. D., R. B. Hutt & B. A. Butt. 1973. Field dispersal of laboratory reared fertile female codling moths. *Environ. Entomol.* 1: 66-69.

White, L. D., F. Proshold, G. G. Holt, K. D. Mantey & R. B. Hutt. 1975. Codling moth: mating and sperm transfer in females paired with irradiated and unirradiated males. *Ann. Entomol. Soc. Amer.* 68(5): 859-862.

Wiesmann, R. 1935. Untersuchungen über den weiblichen Genitalapparat, das Ei und die Embryonalentwicklung des Apfelwicklers, *Carpocapsa (Cydia) pomonella*. *Mitt. Schweiz. Entomol. Ges.* 16: 370-377.

Wildbolz, Th. 1958. Über die Orientierung des Apfelwicklers bei der Eiablage. *Mitt. Schweiz. Entomol. Ges.* 31: 25-54.

Williams, D. G. & G. McDonald. 1982. The duration and number of immature stages of codling moth *Cydia pomonella* (L.) (Tortricidae: Lepidoptera). *J. Aust. Entomol.* 21: 1-4.

Witehead, F. E. 1944. Variations of codling moth infestations in apple varieties. *Proc. Oklahoma Acad. of Sci.* for 1944: 30-33.

Wong, T. T. Y., M. L. Cleveland, D. F. Ralston & D. G. Davis. 1971. Time of sexual activity of codling moth, *Laspeyresia pomonella* (L.), in the field. *J. Econ. Entomol.* 64: 553-554.

Wood, T. G. 1965. Field observations on flight and oviposition of codling moth and mortality of eggs and first-instar larvae in an integrated control orchard. *N. Z. Agric. Res. J.* 8: 1043-1059.

Woodside, A. M. 1944. Codling moth infestation at different heights in apple trees. *Virginia Agricultural Experiments Station Bulletin.* 360.

Worthley, H. N. 1932. Studies of codling moth flight. *J. Econ. Entomol.* 32: 559-565.

Wyniger, R. 1956. Über die Wirkung von abiotischen Faktoren auf die Entwicklungsvorgänge in Apfelwicklerei. *Mitt. Schweiz. Entomol. Ges.* 29: 41-57.

Yokoyama, V. Y. & G. T. Miller. 1988. Laboratory evaluations of codling moth (Lepidoptera: Tortricidae) oviposition on three species of stone fruit grown in California. *J. Econ. Entomol.* 81(2): 568-572.

Yokoyama, V. Y. & G. T. Miller. 1988. Laboratory evaluations of oriental fruit moth (Lepidoptera: Tortricidae) oviposition and larval survival on five species of stone fruits. *Econ. Entomol.* 81(3): 867-872.

Yothers, M. A. 1927. Summary of three years' tests of trap baits for capturing the codling moth. J. Econ. Entomol. 20: 567-575.

Zech, E. 1955. Die Flugzeiten des Apfelwicklers im Jahre 1954 und der Flugverlauf während der Abende und Nächte. Nachrbl. f. Deutsch. Pflanzenschutzd. 2: 29-35.

Zohren, E. 1968. Laboruntersuchungen zu Massenzucht, Lebensweise, Eiablage-verhalten der Kohlfliege. *Chotophila brassicae* Bouche (Diptera, Anthomyiidae). Z. ang. Entomol. 62: 139-188.