SURVIVAL OF EGG, NEONATE AND WANDERING-PHASE LARVAE OF THE INDIANMEAL MOTH (*PLODIA INTERPUNCTELLA* (HÜBNER)) EXPOSED TO SURFACE AND AEROSOL APPLICATIONS OF METHOPRENE

by

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B.S., University of Wyoming, 2001 M.Ag., Oklahoma State University, 2004

AN ABSTRACT OF A DISSERTATION

Submitted in partial fulfillment of the requirements for the degree DOCTOR OF PHILOSOPHY

Department of Entomology

College of Agriculture

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Abstract

The Indianmeal moth, *Plodia interpunctella* Hübner, is capable of infesting a number of different commodities including a wide variety of grains, nuts and finished stored products. Therefore, control of the Indianmeal moth is especially needed in areas where food is being stored for human consumption. Increased concerns of consumers and producers regarding the impact of conventional insecticides on the environment and on human health has prompted scientists and the agricultural chemical industry to search for insecticides that do not affect mammalian nervous systems and have limited effects on non-target organisms. One group of insecticides with reduced risks is insect growth regulators (IGRs), which are substances that mimic insect hormones essential to normal development and reproduction.

Currently methoprene, a juvenile hormone analog, is labeled for direct application to stored grains, as well as a contact insecticide and as an aerosol application inside mills, warehouses, and indoor food storage facilities. Surface treatments and aerosol space applications can be effective ways to treat the interior surfaces and storage areas of warehouses and food processing facilities. There is little recent research with large-scale aerosol applications in storage sites; furthermore, there are no published references in the scientific literature regarding efficacy of using methoprene alone in aerosol form. Therefore, the purpose of this research was to evaluate the use of surface and aerosol applications of methoprene on finished stored-product packaging materials and facilities for the control of *P. interpunctella*.

Results of this research showed that while methoprene has good residual activity, and efficacy is unaffected by temperature, surface applications of methoprene on packaging materials is not as effective for control of *P. interpunctella* as aerosol applications of methoprene. Aerosol methoprene is highly effective alone and in combination with conventional chemicals for control of eggs and wandering-phase larvae. Simulations with a population growth model make it possible to estimate impact of insecticide treatments at different temperatures and application times on populations of *P. interpunctella*. Aerosol treatments are also economically viable as part of an overall integrated pest management program.

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Literature Review

Biology, Significance and Control of the Indianmeal moth

Life History Characteristics

The Indianmeal moth, *Plodia interpunctella* Hübner, is a widespread pest of stored products which infests a number of different commodities (Tzanakakis 1959, Sedlacek et al. 1996, Arbogast 2007a). Larvae consume a wide variety of nuts, grains, and finished stored products, and have been reported or collected from most areas of the world (Tzanakakis 1959, Rees 2004, Arbogast 2007b). This pyralid moth may be distinguished from other members of the family by the characteristic markings on its forewings: reddish-brown on the outer two-thirds, whitish-grey on the inner third (USDA 1986).

The adult females typically lay 100-300 white to grayish eggs, singly or in groups, on or near a food source (USDA 1986). The number of days until eggs hatch depends primarily on temperature (Bell 1975, Sedlacek et al. 1996, Johnson 2007). After hatching, the neonates disperse in search of food; their small size enables them to enter containers that have even minute openings due to manufacturing defects or mechanical damage. Larvae are external feeders with the ability to bore into some package materials. The duration of the larval period (five molts) depends on environmental factors such as temperature and diet (Tzanakakis 1959, Mohandass et al. 2007). The larvae range in color from tan to light green to pale pink and can vary in size depending on diet and population size. Once larvae reach the 5th instar or wandering phase, they typically leave the food medium in search of a pupation site (Sedlacek et al. 1996).

The developmental time from egg to adult varies with temperature, relative humidity, and diet (Bell 1975, Mbata and Osuji 1983, Johnson et al. 1992, Subramanyam and Hagstrum 1993, Arbogast and Chini 2005) and the life cycle may be extended considerably by entering diapause (Tzanakakis 1959, Mohandass et al. 2007). Typically, the time from egg to adult is approximately 27 days at 32.2°C (90°F) (Jenson, unpublished data). At this developmental rate, a population infesting a commodity could have as many as eleven generations per year in the presence of other favorable growing conditions. The Indianmeal moth's short life cycle,

combined with its high reproductive capacity, give it the potential for significant product damage in and around food storage facilities.

Significance: Damage Caused by Indianmeal moth

Indianmeal moth can be problematic in all stages of the production of food products from the grain bin to the end user. The list of commodities that the Indianmeal moth is known to infest is long and extensive and includes many different grains as well as dried fruit and nuts (Mohandass et al. 2007). Infestations lead to equipment damage and direct losses in raw and finished products. They are also unsightly and have an unpleasant odor, which causes negative consumer feed-back in addition to the problems they create during the food manufacturing process.

Beginning with the on-farm storage of a commodity, populations of Indianmeal moth in grain bins can cause heat and moisture gradients resulting in bacterial or fungal growth, in addition to the physical feeding damage which reduces grain weight. Unmanaged infestations can then be transported with the commodity, or the product can become infested during transit. In the absence of an in-bound inspection or sampling program, Indianmeal moth can enter a facility unnoticed along with the raw product. Adults are strong fliers and can enter facilities through open doors and windows. Indianmeal moth can also be a problem in facilities that store finished stored products; in manufacturing plants; storage warehouses or even retail stores.

Early instars are able to penetrate packaged products that have only tiny ruptures or seams, even when it appears that packages are tightly sealed (Mullen 1994, Mowery et al. 2004). Larvae are only limited by distance from the resource and size of their head capsule. Young larvae typically stay in the food, while mature larvae seek concealed pupation sites, therefore they may be difficult to detect in homes and food storage facilities.

Damage caused by Indianmeal moth includes losses from direct feeding, product contamination and creation of favorable conditions for mold and bacterial growth. Aside from the regulations concerning insect parts and frass in food for human consumption for which infestation is a problem, the sticky silken webbing that all larval stages produce can be just as harmful and add to the overall damage caused by these insects (Tzanakakis 1959). For example, extensive webbing can cause food particles to clump, binding and slowing of equipment, or directly render the product unmarketable. Therefore, control of the Indianmeal moth is especially needed in areas where food is being stored for human consumption.

Management Options

The late instar wandering-phase of the Indianmeal moth may leave the food source to find a pupation site, thus providing a window for control. However, this larval stage is difficult to control with conventional insecticides. Arthur (1997) reported little susceptibility of 5th instar larvae to residues of deltamethrin dust except during the immediate period after treatment. Cyfluthrin and chlorpyrifos-methyl are only effective at high rates (22-30 ppm) immediately following application (Arthur 1989, 1995). Other pests such as adult red flour beetles, *Tribolium castaneum* (Herbst), and adult merchant beetles *Oryzaephilus mercator* (Fauvel), have much greater sensitivity to chlorpyrifos-methyl than do mature moth larvae (Arthur 1989). The wandering-phase is also much more tolerant of conventional insecticides than are younger Indianmeal moth instars, adult coleopterans (Arthur et al. 2004), or other lepidopteran larvae (Yue et al. 2003).

When eggs are oviposited in areas that may come in contact with a surface or aerosol application of insecticide, this provides another potential window for control. Eggs of stored grain and fruit pests are often the most difficult life stage to kill using conventional fumigants; including methyl bromide (Weller and Morton 2001, Armstrong and Whitehand 2005). They are also relatively heat and cold tolerant (Mahroof and Subramanyam 2006, Johnson 2007). With high rates of insecticide application, low residual activity, and the apparent tolerance of the 5th instars to conventional insecticides, a different type of insecticide is needed to control the Indianmeal moth in areas where processed and packaged foods are stored. Other management options should also be considered.

Basic components of an integrated pest management program for Indianmeal moth are sanitation, chemical and physical control and inspection. Products should be inspected for signs of infestation prior to storage in a warehouse or residence. Removal of product waste and debris in and around facilities will reduce the amount of food resources and pupation sites available for larvae. Many studies have proven the efficacy of using physical control methods such as heat treatments (Johnson et al. 2003, Mahroof and Subramanyam 2006), freezing (Johnson and Wofford 1991, Johnson 2007), vacuum sealing (Mbata et al. 2004) and cool air aeration (Reed and Arthur 2000, Johnson, et al. 2002, Kaliyan et al. 2007). Chemical control methods using sex pheromones for mass capture and mating disruption may also reduce Indianmeal moth populations (Nansen and Phillips 2004, Nansen et al. 2006). Use of conventional insecticides

has been shown to be moderately effective; but there is evidence that insect growth regulators such as methoprene and hydroprene may provide highly effective control for both eggs and larvae (McGregor and Kramer 1975, Fajardo and Morallo-Rejesus 1979, Mohandass et al. 2006b, 2006c). With the scheduled phase-out of methyl bromide (Fields and White 2002), there is a definite need to find alternatives to conventional fumigation.

Chemical Control Using Insect Growth Regulators

Insect Growth Regulators: History and Attributes

Due to growing public and scientific concern about the effects of insecticides in the environment, and the observation of insect resistance to the current insecticides on the market, the search for a "better" insecticide is an important research priority. "Third generation" insecticides, which alter insect growth and development, were discovered as potential insecticides in 1952 with the report of lowered egg hatch in cultures of the linden bug reared with paper towels in the jar (Tunaz 2004). Subsequent studies showed that juvabione, a juvenile hormone mimic, was present in the paper used in rearing (Tunaz 2004). Carrol Williams' group began isolating the "juvenile hormone" in 1956 from the abdomen of male *Cercropia* moths (Roman et al. 1967, Slama 1971, Klowden 2002). Following this major discovery, groups of researchers began to study these compounds for their function in insects and insecticidal properties. Insect growth regulators (IGRs) are substances which act within an insect to accelerate or inhibit a physiological regulatory process essential to the normal development of that insect (Siddall 1976). Since that time a number of studies have examined the effects and potential of IGRs, especially juvenile hormone, on a wide range of insect species.

As reported in Mondal and Parween (2000), there are both advantages and disadvantages to the use of IGRs in the control of insects. Discussions often compare and contrast their use with conventional insecticides such as carbamates and organophosphates. The primary difference between IGRs and conventional insecticides is that IGRs are considered to be "reduced risk" insecticides for humans and other non-target organisms with high selectivity, whereas conventional insecticides typically have low selectivity and substantial risks. "High selectivity" refers to the physiological sensitivity of the target organism compared with other non-target organisms. Insecticides that have high selectivity are generally considered "safer" due to their reduced risks of detrimental affects to non-target organisms such as birds and

mammals. The "reduced risk" of using IGRs can, in part, be attributed to the specific life stage that is being targeted in the pest, relatively quick degradation in the environment, and low mammalian toxicity (Mondal and Parween 2000). In addition to these characteristics, reduced risk also refers to other negative affects such as groundwater contamination and resistance development. Lower risk to non-target species is especially advantageous in controlling insects associated with human food.

Another advantage of IGRs is their rapid degradation in the outside environment due to UV radiation and short half-lives. However, in closed, often dark conditions where commodities are stored, IGRs can persist longer than in the outside environment. Regardless, there is evidence that IGRs do not change the quality of stored products (Mondal and Parween 2000). IGRs have been shown to work in "low" doses compared to conventional insecticides and have especially important applications in stored product situations. Along with low risk, high selectivity and low persistence, IGRs are relatively stable over a variety of temperature and relative humidity conditions. Because of the nature of these compounds, the use of insect growth regulators seems to fit with existing integrated pest management programs (Tunaz 2004).

Among the disadvantages of IGRs is that they work relatively slowly. For example, hormone mimics can prolong larval life, leading to a longer window in which the larvae can feed (Mohandass et al. 2006c). By comparison, conventional insecticides typically cause rapid knockdown and nearly immediate mortality of pest populations. IGRs also have lower contact mortality than conventional insecticides; instead, their primary effect is to reduce population growth by not allowing adult emergence, or sometimes by causing impairment to adults following contact with IGRs in the larval stages. While relative risk to humans and other mammals is quite low, these compounds can negatively affect beneficial insects in the same way as pest species; this can be either an advantage or a disadvantage depending on the system that is being targeted. Timing of application may also be a problem because some IGRs only work in certain developmental stages. Many IGRs are most effective when they can be ingested or readily absorbed through the cuticle prior to molting. Therefore some life stages, such as pupal Lepidoptera, and most adult insects are not affected by applications of these types of chemicals. Another disadvantage of IGRs is that very little is known about the sub-lethal effects of these types of chemicals in field situations. It is not known how significantly exposure to affects reproduction and population growth in pest species.

Chemistry of Juvenile Hormones and their Analogs

Juvenile hormone (JH) was initially described as an "inhibitory hormone" because it prevented metamorphosis to the adult stage in *Rhodnius prolixus* in experiments conducted by Wigglesworth (Klowden 2002). Since that time, researchers have found that JH has many functions in insect sexual behavior and egg production, embryonic development, metamorphosis, migration, diapause and social structure in eusocial insects (Hartfelder 2000, Davey 2007). JH is a sesquiterpene with several forms produced in the corpora allata (Chapman 1998, Minakuchi and Riddiford 2006). These forms (JH0, JHI, 4-methyl JHI, JHII, JHIII, and JHIII bisepoxide), which differ slightly in regard to their carbon number, vary by order, with most insects having the most primitive form, JHlll (Klowden 2002). Because JH is such an important hormone with many roles in insects, it makes it an ideal target for management of insect pests.

JH modifies the insect's response to ecdysteroids and prevents change in commitment of epidermal cells (Chapman 1998, Klowden 2002, Truman and Riddiford 2002). During the larval stages of insects, the ratio of JH to ecdysteroid is high until prior to ecdysis, when levels of JH esterase and ecdysteroid rise. In this way, JH works as a "status quo" hormone by suppressing the developmental pathways to new cuticle production, effectively 'keeping' the insect 'young'. When JH esterase and JH epoxide hydrolase degrade JH in the hemolymph, ecdysteroid is allowed to act on epidermal cells to initiate metamorphosis (Chapman 1998, Klowden 2002, Minakuchi and Riddiford 2006). One mode of action of juvenile hormone mimics and related IGRs is to disrupt the endocrine system and metamorphosis by introducing JH-like compounds that prevent eclosion to the pupal and adult stages. Suspending insects in the larval stages or introduction of supernumerary molts prevents them from becoming reproductively mature, effectively reducing pest populations.

Though insect resistance to hormone mimics was thought to be impossible at one time, like all insecticides, insects can become resistant to these compounds and cross-resistance may a problem (Dhadialla et al. 1998, Mondal and Parween 2000). Insect resistance to methoprene, hydroprene, kinoprene, pyriproxifen, RH 5992 and diflubenzuron have been reported (Hoffmann and Lorenz 1998). The current theories of resistance are placed in three broad groups: resistance caused by the mutation or impaired function of a gene or JH receptor; resistance due to increased metabolic degradation caused by esterases; and resistance due to the action of microsomal P450

enzyme (Feyereisen 1999, Browder et al. 2001, Truman and Riddiford 2002). In Drosophila (Diptera) there is a *Methoprene-tolerant (Met)* gene which belongs to the bHLH family of transcriptional regulators (Feyereisen 1998, Truman and Riddiford 2002). The presence of this gene in methoprene-resistant lab colonies was confirmed and expression results in tolerance to JH mimic insecticides and sub-lethal effects, such as bristle malformation and malrotated genitalia in male flies (Wilson et al. 2006). A similar gene was also found in *Tribolium* (Coleoptera) (Konopova and Jindra 2007). Juvenile hormone esterase is vital in the metamorphosis of insects; both production and degradation of juvenile hormone is important and fluctuates throughout the insect life cycle (Klowden 2002). Resistance to JH mimics could be associated with activation of the esterases and mixed function oxidases. Another possibility is that microsomal P450 is involved in the metabolic degradation of JH mimic insecticides and also in insect resistance to these toxicants. The proposed mechanism of resistance related to this enzyme system is that detoxification is occurring and results in a block of JH synthesis, leading to adult sterility and precocious adults (Feyereisen 1999). Overall, the benefits of using an insect's own hormone to safely control pest populations must be weighed against the non-target and environmental effects, as well as the potential for resistance development.

Commercial Availability of JHA

There are many juvenile hormone analogues (JHA) and juvenile hormone mimics that are used as insecticides. These include S-hydroprene (Ethyl(2E,4E,7S)-trimethyl-2,4-dodecadienoate), methoprene (Isopropyl (2E,4E)-11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate, and S-kinoprene (2-Propynly(s-(2E,4E))-3,7,11-trimethyl-2,4-dodecadienoate) compounds. Again, these compounds have different effects depending on exposure interval and life stage, and they are mostly used as contact poisons (Mondal and Parween 2000, Mohandass et al. 2006a). Hydroprene is labeled for direct use as a surface treatment in food storage facilities, and as a vapor strip in cabinets and cupboards. Methoprene is labeled for a variety of surface treatments indoors and outdoors and has action against many insects such as flies, mosquitoes, ants, moths, beetles, and mites (Henrick 2007). It is effective on the lesser grain borer, *Rhyzopertha dominica* (F.), a major internal feeder on stored grains (Arthur 2004, Chanbang et al. 2007). Female lesser grain borer oviposit eggs on the outside of a grain kernel, and after hatching the first instar bores into the grain. During this brief window, the insect can be exposed to methoprene residues on the grain surface. Currently methoprene is labeled for direct

application to stored grains, as well as a liquid or aerosol application inside mills, warehouses, and indoor food storage facilities (Central Sciences International 2002, 2004).

Methoprene and Stored Product Protection

Several studies have been published on the incorporation of methoprene into the pest's food source (Strong and Diekman 1973, McGregor and Kramer 1975, Loschiavo 1976, Firstenberg and Silhacek 1976, Mian and Mulla 1982, Manzelli 1982). Because of the occurrence of favorable conditions for both pests and use of IGRs in storage situations, it is no surprise that there has been extensive research conducted in this area. Methoprene incorporated into food media has been shown to reduce survival of Indianmeal moth and several other stored-product pests with increasing concentrations (Loschiavo 1975, 1976; McGregor and Kramer 1975, Strong and Diekman 1973). Even at very low doses of 2-5 ppm, survival to the adult stage was greatly diminished (McGregor and Kramer 1975, Loschiavo 1976, Fajardo and Morallo-Rejesus 1979). Also of interest are the sub-lethal effects of methoprene, such as deformation, supernumery molts, sterility and reduced longevity of adults, which may help to control populations of the pest (Fajardo and Morallo-Rejesus 1979). Although there has been considerable research with IGRs in stored products, they have received increased attention for incorporation into a variety of insect pest management programs (Oberlander et al. 1997, Mondal and Parween 2000, Mohandas et al. 2006a).

Factors Affecting Efficacy of Methoprene for Control of Indianmeal Moth

Temperature

The number of days required for Indianmeal moth development is influenced by a number of factors, including diet (Johnson et al. 1992), relative humidity (Bell 1975), and temperature (Cline 1970). Temperature can heavily influence the number of eggs laid as well as the rate of development in the presence of other satisfactory conditions (Tzanakakis 1959, Mohandass et al. 2007). Short exposures to low temperatures (2.4°C) in the egg stage have been shown to decrease survival (Cline 1970), while rearing insects at high temperatures (35°C) has been shown to decrease the ability to reproduce (Johnson et al. 1992). Bell (1975) reported the range of suitable temperatures as 15 to 30°C for development and reproduction of the Indianmeal moth. Development is also affected by relative humidity, diet, and natural variation of different

moth populations. Temperature can also influence the effectiveness of the insecticides. Toxicity of organophosphates generally increases as temperature increases, while toxicity of pyrethroids can decrease with temperature (Arthur et al. 2004). This interaction between temperature and toxicity can have a profound effect on the performance and efficacy of a particular insecticide, and also on the target insect species. This influence becomes especially important when methoprene is applied in combination with a chemical that has reduced efficacy with varying temperatures.

Packaging and Surface Types

One possible way to reduce insect infestation by companies supplying finished products to the consumer is the investment in "better" package design. This can be prompted by consumer complaints about insect damaged products, packages damaged in shipment, or the return of damaged products to the manufacturer. Although the manufacturer may not be directly responsible for the damage done to packaging in shipment, the company still remains accountable for the finished products (Highland 1978). Economic losses as a result of insect infestation in finished products exceed the loss of quantity and quality that is easily quantified, but can extend to loss of consumer confidence, ultimately tarnishing the company's image in the eye of the consumer.

Insects can enter a finished product along one of two routes, chewing or boring into the package, or entering a rupture in the package that was initially caused by something else. Indianmeal moth larvae are capable of boring into many common package types (Cline 1978, Highland et al. 1984). There are many ways to decrease the incidence of insects entering finished product packages, including the use of repellents, odor barriers, insecticides and insect-resistant packaging (Highland 1978). Choices in package design, such as selection of material type and method of sealing, can reduce insect entry. Other modifications such as type of shipping containers may make insect entry difficult to impossible. The use of methoprene on finished product packaging materials may not prevent entry into the package, but could suppress infestation by preventing subsequent generations from infesting other packages in the shipment or warehouse.

Application Method

As stated previously, the wandering-phase larva of the Indianmeal moth is difficult to control using conventional insecticides. Methoprene has been shown to be effective against this species when incorporated into food sources, and incorporation of methoprene into packaging materials could provide another source of protection for stored products and storage facilities, without the risks associated with conventional insecticides. Application of the insecticide to packaging materials instead of incorporation into the product may also reduce the need for precise timing and multiple applications. Food packages are designed to protect food from the manufacturer until the product reaches the consumer. Infestations by insects can occur during the manufacturing process, enroute to the outlet, and even on the grocery shelves (Mullen and Mowery 2003). Infestations reduce consumer confidence in the product, so elimination of infestations is vital. Several packaging methods are available, including insect-resistant packaging (physical barrier), repellent treatments, and odor barriers (Mullen 1994) however, there are few packages that can deliver protection for all products in all situations (Mullen and Mowery 2003). Direct application of methoprene to the food packaging materials, or incorporation of methoprene into the package coating, may be a useful tactic to reducing warehouse and storage facility infestations. However, there are no previous reports on the efficacy against Indianmeal moth larvae of methoprene applied to packaging materials. Package and surface materials, on which methoprene is applied, may also influence the efficacy of this insecticide.

Aerosol space applications may also be an effective way to treat the interior surfaces and storage areas of warehouses and food processing facilities. Recent field studies by Arthur (2008) have shown that aerosol applications of pyrethrin can control the red flour beetle, *Tribolium castaneum* (Herbst). However, there are no published references in the scientific literature regarding efficacy of using methoprene alone in aerosol form to control the Indianmeal moth. A common pest management strategy is to use methoprene in combination with a pyrethrin or pyrethroid insecticide. Systems for ultra low volume (ULV) aerosol delivery have been designed and installed in commercial milling and storage facilities.

Mixtures of Insecticides

Currently, in manufacturing and food processing plants where aerosol fogging systems are installed, pest managers are using conventional insecticides alone, and in combination with

methoprene. Efficacy of these chemicals individually, and in combination, needs to be evaluated. Therefore, the current research is being undertaken to evaluate the use of methoprene on finished stored-product packaging materials and surfaces for the control of Indianmeal moth.

Predicting How Methoprene Will Perform in Real-World Situations

Population Growth Models

Many ecological models have been developed for stored product pests (Throne 1995) and can be adapted to different systems. Models for the almond moth, *Cadra cautella* (Walker), the predator *Lyctocoris campestris* (F.), the flat grain beetle, *Cryptolestes pusillus* (Schoenherr) and the red flour beetle, *Tribolium castaneum* (Herbst), have been developed (Throne 1995). As far back as 1967, computers have been used to simulate population development (Throne 1995). Many factors influence development of stored product pests; temperature, relative humidity and diet are all important factors to be included in any of these models (Throne et al.1998). Once a model is developed to predict growth rates on one type of diet, it can be modified to show rates of growth after chemical application or other types of diets.

Economic Models

An economic analysis can compare different application methods and rates of methoprene alone, and in combination with conventional insecticides, to enable food production plant managers and warehouse managers to make better decisions. Integrated pest management in field crops has a long history of using concepts such as economic injury levels and thresholds to determine timing of control strategies. With extremely low thresholds in finished stored product situations, a slightly different approach is needed (Higley and Wintersteen 1992, Stejskal 2002, 2003). Many types of economic analysis have already been applied to other systems, including field crops and ornamentals (Headley and Hoy 1987, Jetter et al. 1997) and grain bins (Tilley et al. 2007) however, the warehouse environment is a novel use of these standard methodologies. These types of analyses will show how methoprene application as a control strategy may be optimized in warehouse environments.

Economic analysis for some other methyl bromide alternatives for specific field crops have been developed using enterprise budgets (Nelson 1996, Byrd et al. 2006). Partial budget analysis, which compares costs of control strategies can be used to determine levels of risk

associated with each strategy (Boehlje 1984). Cost inputs in the partial budget in this analysis for a stored product situation could be chemical, applicator and/or equipment, shutdown/loss of production, labor and sampling (and costs associated with this process).

Conducting an analysis of cost and risk associated with different control strategies can determine whether methoprene is an economically-competitive control strategy compared with other methods such as sanitation, conventional insecticides or physical control methods. An analysis can also determine which type of methoprene application is the most effective and efficient strategy. A cost versus effectiveness tradeoff model can be used if the partial budget analysis reveals significant differences between these different systems. Aerosol methoprene may be an economically viable alternative to methyl bromide and costly fumigations.

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Objectives

The overall hypothesis I wish to test is that methoprene on packaging materials offers an effective control strategy against Indianmeal moth. The specific objectives of this study are to:

- 1. Examine the effect of exposing egg-neonate and 5th instar Indianmeal moths to methoprene on paper packaging materials under different temperatures on survival to the adult stage.
- 2. Test the effect of different types of packaging and surface materials on the efficacy of methoprene on survival of the Indianmeal moth to the adult stage.
- 3. Determine the effect of aerosol application of methoprene alone and in combination with an esfenvalerate insecticide on the survival of Indianmeal moth to the adult stage.
- 4. Compare the survival of Indianmeal moths to the adult stage on product packaging, diet, and direct exposure to methoprene and synergized pyrethrin aerosols in a field situation.
- 5. Simulate population dynamics of Indianmeal moth using responses to methoprene and combinations of methoprene and other conventional insecticide treatments with time and temperature as variables.
- 6. Develop an economic model to determine costs and risk associated with methoprene and other chemical applications to control Indianmeal moth.

CHAPTER 1 - Efficacy of Methoprene Applied at Different
Temperatures and Rates to Different Surface Substrates to
Control Eggs and Fifth Instars of Indianmeal moth (*Plodia*interpunctella Hübner)

Abstract

As regulations on use of insecticides continue to become more stringent, there is a need to look for lower impact alternatives, including insect growth regulators. The insect growth regulator, methoprene, has been shown to decrease populations of Indianmeal moth (*Plodia interpunctella* Hübner) by preventing development to the adult stage. Eggs were exposed to methoprene on paper packaging materials treated at the label rate for surface application then allowed to mature to adulthood at four temperatures. Larvae were exposed for different intervals ranging from 0-4 hours on a paper packaging material treated at the several rates of methoprene, at temperatures ranging from 20 to 32°C. Survival of larvae to the adult stage decreased with increasing exposure and rate of insecticide. Eggs and larvae were exposed in a similar manner to methoprene on nine different surface and packaging materials. While temperature did not affect efficacy of methoprene, surface materials did influence rate of survival to the adult stage. Results show that the surface substrate affects control of the Indianmeal moth through the use of methoprene.

Keywords: temperature, methoprene, packaging materials, Indianmeal moth

Introduction

Insect growth regulators may fill a critical need for lower-impact insecticides in food processing facilities (Mondall and Parween 2000, Mohandass et al. 2006a). The scheduled phase-out of methyl bromide, coupled with consumer demand for safer food products, makes insect growth regulators an attractive management option (Campbell et al. 2004, Mondall and Parween 2000). The insect growth regulator (IGR), methoprene, is a juvenile hormone analog that has many biological effects and works as an insecticide to reduce pest populations by preventing maturation to the adult stage (Oberlander et al. 1997, Henrick 2007). Specifically, there have been many studies demonstrating effectiveness of methoprene incorporated into an insect's food source (Strong and Diekman 1973, McGregor and Kramer 1975, Loschiavo 1976, Firstenberg and Silhacek 1976, Mian and Mulla 1982, Manzelli 1982). Methoprene is currently labeled for direct application to stored grains, as well as a liquid space application inside mills, warehouses, and indoor food storage facilities (Central Sciences International 2002, 2004).

The Indianmeal moth (*Plodia interpunctella* Hübner) is a serious pest as it occurs on many commodities and stored products and occurs during the entire food manufacturing process, from the grain bin to the table (Doud and Phillips 2000, Johnson et al. 2003, Mahroof and Subramanyam 2006). The number of days required for Indianmeal moth development (and subsequent population dynamics) is influenced by a number of factors, including diet (Johnson et al. 1992), relative humidity (Bell 1975), and temperature (Cline 1970). Temperature can heavily influence the number of eggs laid as well as the rate of growth and development in the presence of other satisfactory conditions (Tzanakakis 1959, Mohandass et al. 2007). Temperature can also influence the effectiveness of some insecticides. Toxicity of organophosphates generally increases as temperature increases, while toxicity of pyrethroids can decrease with temperature (Arthur et al. 2004). This interaction between temperature and toxicity can have an important impact on efficacy of a particular insecticide. The Indianmeal moth's short life cycle, combined with its high reproductive capacity, give it the potential for significant product damage in and around food storage facilities. For these reasons, there is a need to assess how temperature affects efficacy of methoprene for control of this pest.

While incorporation of methoprene into a pest's food source can be highly effective, incorporation onto packaging materials could provide another source of protection for stored

products and storage facilities, without the risks associated with conventional insecticides. Application of the insecticide to packaging materials instead of incorporation into the product may also reduce the need for precise timing and multiple applications. Food packages are designed to protect food from the manufacturer until the product reaches the consumer. However, infestations by insects can occur during the manufacturing process, enroute to the outlet, and even on the grocery shelves (Mullen and Mowery 2003). Infestations reduce consumer confidence in the product, so elimination of infestations is vital. Due to the occurrence of favorable conditions for both pests and use of IGRs in food storage situations, there has been considerable research conducted in this area. However, few studies have demonstrated efficacy against the Indianmeal moth or its efficacy when used as a surface or packaging treatment.

The Indianmeal moth is difficult to control with conventional insecticides, especially in the late-larval stages (Arthur 1997). Cyfluthrin and chlorpyrifos-methyl are only effective at high rates (22-30 ppm) immediately following application (Arthur 1989, 1995). Eggs of stored grain and fruit pests are often the most difficult life stage to kill using conventional fumigants, even methyl bromide (Weller and Morton 2001, Armstrong and Whitehand 2005). Eggs of the Indianmeal moth are also relatively heat-and cold-tolerant, making this life stage an important target for control of populations (Mahroof and Subramanyam 2006, Johnson 2007). The late-instar wandering phase larvae may leave the food source to find a pupation site, thus providing one window for control. When eggs are oviposited on packaging, shelving, flooring, or on spilled products, the eggs may come into contact with a surface application of insecticide; thus, eggs provide a second window for control. Therefore, the objectives of this study were to determine 1) whether temperature affects efficacy of methoprene in preventing survival to the adult stage for exposed eggs and larvae, and 2) if surface type modifies the effectiveness of methoprene.

Materials and Methods

General Procedures

The Indianmeal moth population that was used in all experiments derived from a laboratory colony established in June 1988 from individuals collected in Riley County, KS, U.S.A., and periodically supplemented with wild-type individuals captured in Riley County. This colony has been maintained on an enriched wheat diet of cracked wheat and shorts (4.4 kg),

brewer's yeast (22g), sorbic and benzoic acid (9.5g each), honey (240ml), glycerin (240ml) and water (120ml) inside environmental growth chambers (Forma-Scientific, Thermo Electron Corporation, Waltham, MA) at 27 ± 1 °C, approximately 50% RH., and in darkness (L:D = 0:24 hrs) at the United States Department of Agriculture Agricultural Research Service (USDA-ARS) Grain Marketing and Production Research Center in Manhattan, KS. Voucher specimens have been deposited in the Kansas State University Museum of Entomological and Prairie Arthropod Research under Lot Number 208. Experiments were conducted in environmental chambers under the same physical conditions as the colony. Humidity chambers were constructed for the temperature experiment using acrylic boxes with lids that contained a saturated solution of distilled water and NaBr, which maintains a constant 57% relative humidity (Greenspan 1977). A more detailed set of procedures for construction and use of these boxes was described by Arthur (2000). The units were approximately 26 x 36.5 x 15 cm rectangular plastic boxes with a waffle-style plastic grid cut to fit the bottom. Separate humidity chambers were used for the various methoprene treatments and the untreated controls. Humidity boxes and environmental chambers were selected randomly for each experiment and re-randomized between blocks of each experiment. Temperature was monitored using digital thermometers checked frequently. Temperature and relative humidity were also recorded using a HOBO data logger (Onset Computer Corporation, Bourne, MA) placed in humidity chambers.

Experiment 1: Survival of Eggs Exposed to Methoprene at Different Temperatures

Eggs and neonates were tested at five temperatures (16, 20, 24, 28, and $32 \pm 1^{\circ}\text{C}$), a range that will support Indianmeal moth population growth. Treatment arenas were constructed by gluing a 100 mm Petri dish inside of a 150 mm Petri dish with industrial adhesive. Approximately 75 g of standard wheat diet was placed uniformly into the outer ring. Preliminary tests show that the amount of diet in each arena is sufficient for normal development to the adult stage of 100 Indianmeal moth eggs. Lids on the 150 mm Petri dishes were altered to have a 5.08 cm (2 in) diameter vent cut in the top with a standard filter paper secured with a non-toxic adhesive to allow air movement. The inner dish had a diameter of 62 cm². The construction of these treatment arenas allowed for movement between the smaller and larger dish. Eggs were placed in the inner dish for testing purposes and after hatch were allowed to exit the inner circle to the diet area. Adult female moths, less than 48-hours-old, were placed in a jar for oviposition

and eggs were collected for a period of three hours. One-hundred 0-3-hour-old Indianmeal moth eggs were counted using an aspirator with a small glass collection jar and placed in the center of treated or untreated papers cut to fit the smaller dish.

The treated and untreated material used in all temperature experiments was uncoated brown Kraft paper similar to that used by the United States Agency for International Development (USAID) for commodity shipments. It was obtained from the Smurfit-Stone Company, Kansas City, MO in 2005. Chosen for its widespread industry use, preliminary experiments indicated that this paper was a suitable material. The methoprene used in experiments was the formulation, Diacon II[®], which is an emulsifiable concentrate of 33.6% active ingredient, 300 grams per liter. This was obtained from Central Sciences International Dallas, TX. Methoprene was mixed with distilled water to achieve the desired concentration. Four concentrations were used: 0 rate methoprene (control), the label rate for surface application (300 mg AI /94m²), 0.5 times the label rate for surface application, and 1.5 times the label rate for surface application. Controls were treated with distilled water alone. Brown Kraft paper was cut into 19 x 13 cm pieces and these were individually treated by dripping 1 ml of one of the test solutions onto the paper and then spreading the liquid over the surface using a glass rod. Dried treated papers were cut into 62 cm² circles to fit inside the smaller dish in the treatment arenas and eggs added. The plates were sealed with Parafilm® and placed into humidity boxes (separate for treatments and controls) inside growth chambers at the different temperatures.

Preliminary observations indicated that 2 to 14 days are required for egg hatch, depending on temperature. After sufficient time for hatching had elapsed, the plates were checked (by unsealing the dishes), and unhatched eggs counted using a microscope. Treated and control papers were removed from the plates when at least 75% of the eggs were hatched in each plate to prevent the larvae from coming into contact with the treated surface after their initial exposure. Days until 75% hatch, first date of adult emergence, the number of days to beginning and ending adult emergence, and total number of adults were recorded and analyzed. Once adult emergence began, treatment arenas were monitored daily until emergence in each plate reached zero. The experimental design was a split plot with temperature (growth chamber) representing the whole plot treatment and methoprene treatment serving as the sub-plot, organized in two randomized complete blocks with four replications each.

Experiment 2: Survival of Fifth Instars Exposed to Methoprene at Different Temperatures

To test the effect of temperature and methoprene exposure to 5th instar larvae on survival to the adult stage, an experiment was conducted similar to that described above for eggs and neonates except that only four temperatures were used: 20, 24, 28, and 32+1°C. Data from the egg-neonate study (Experiment 1) showed that 16°C is near the minimum temperature needed for survival of the test species as survival at that temperature was very low. Therefore, this temperature was omitted. An additional difference in the experimental design was that only two methoprene treatments were used – either the label rate or an untreated control. Preliminary studies suggest short exposure intervals will be sufficient for control because wandering phase (5th instar) Indianmeal moth larvae are very susceptible to IGRs (Mohandas et al. 2006b;c). Kraft paper was cut and treated in the same manner as previously described; however, after the paper dried, it was not cut. The paper was folded into packets that allowed for larval movement inside and then secured with painter's tape at the seams in a manner similar to grocery bags. Activelywandering larvae were collected using a soft forceps, and placed into these packets for the duration of the exposure interval. For each treatment, ten 5th instar larvae were exposed for 0, 0.5, 1, or 2 hours. After the exposure period, packets were opened and remaining larvae (minus those damaged or escaped) were transferred to 150 mm Petri dishes and allowed to mature to the adult stage. These dishes had filter paper vents, as described above, and were sealed with Parafilm® and placed in humidity boxes inside incubators of different temperatures.

In the second portion of this experiment (which I will refer to as experiment B, with the former portion designated as experiment A), four concentrations (0.5x the label rate, the label rate, 1.5x the label rate, and a control) of methoprene were used and another exposure interval (4 hours) was added. The additional exposure interval was deemed necessary to demonstrate increased mortality due to exposure time based on results from the previous experiment. The number of temperatures was reduced to three: 22, 27 and $32 \pm 1^{\circ}$ C. All other procedures were the same, including the construction and use of the paper packets. Data collected for both temperature experiments included the number of days until first adult emergence and the overall number of emerging adults. The experimental design was a split-plot with the same treatment structure as the egg and temperature experiments. In the first experiment, temperature represented the whole plot treatment and length of exposure interval the sub-plot treatment. In

the second experiment, concentration*exposure interval represented the sub-plot treatment. Treatments were organized into randomized complete blocks and three replications per experiment were completed.

Data for all temperature experiments (both eggs and larvae) were analyzed using the MIXED procedure (PROC MIXED) of the Statistical Analysis System (SAS) (SAS Institute 2001). For percent survival to the adult stage for each life stage, means and standard errors were calculated using the MEANS Procedure of SAS. Means for treatments were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.

Experiment 3: Survival of Eggs Exposed to Methoprene on Different Surfaces

In this experiment, temperature was fixed at $27\pm1^{\circ}$ C and methoprene concentration was the label rate for surface applications with distilled water controls. To test the effect of packaging type on eggs, tests were conducted using similar procedures and the same test arenas as described above in the egg/temperature experiments. Nine package and surface types were examined: uncoated brown Kraft paper, black paper with a laminated coating, cardboard, cardboard with a laminate coating, plastic pallet wrap, Kraft paper with a waxy coating, concrete wood and tile. Brown Kraft paper is described above. Cardboard pet food cartons (for laminated and uncoated cardboard) and multi-walled pet food bags (for laminated paper and waxy paper), were obtained from Northwest Coatings (Greensboro, NC) in 2006. Package materials were chosen to represent the variety of materials that might be encountered in a typical grocery warehouse. Package materials were cut to a uniform size and treated with methoprene at the label rate using a Badger[©] 100 artist's airbrush (Badger Corporation, Franklin Park, IL) to spray the materials. The airbrush was used instead of the roller method due to the variety of packaging and surface types.

The surface materials (concrete, wood and vinyl floor tile) were chosen as representative of three flooring types in most manufacturing plants and warehouses. Wood (in the case of shelving and stackable pallets) also represents a surface that might be accessed by pests. Surface materials were constructed in a slightly different manner. Concrete treatments were made by pouring a thin layer (10 m) of concrete patching material (Rockkite[®]) in the bottoms of 100 mm Petri dishes. This slurry was water-based and prepared by mixing about 2000 g concrete mix

with 1.0 L of tap water. Construction grade plywood (1.25 cm thick) and self-adhesive vinyl flooring were cut to fit inside the 100 ml Petri dishes to make the individual treatments for those surfaces. Methoprene was sprayed onto surfaces using the same air-brush technique and all treatments were allowed to dry before exposing eggs. After drying, twenty eggs (less than 3 hours after oviposition) were placed in the center of the treatment arenas. Again, after 75% hatch, treated materials were removed from the arenas to prevent further exposure in other life stages. This experimental design was a randomized complete block design conducted in four blocks (replicates) over time. Treatment structure was a 2 x 6 x 3 factorial, with two concentrations (control and insecticide treatment), six packaging types, and three surface material types. Data from each treatment, including total number of emerged adults, total number of days until beginning adult emergence, number of days until adults were finished emerging were collected and analyzed using the same SAS procedures as previously described.

Experiment 4: Survival of Fifth Instars Exposed to Methoprene on Different Surfaces

To test the effect of packaging and surface materials on 5th instars, the same package materials were used as in the egg experiment. Temperature was fixed at 27±1°C and methoprene concentration was the label rate for surface applications with distilled water controls. Packets, as described in the temperature experiments, were made from the packaging materials after insecticide treatment. This experiment was conducted at the same temperature and treatment levels described previously. Because of the wide variety of packaging and surface material types used, treatment arenas were constructed in different manners, but were treated and allowed to dry for the same period of time. Two complete blocks with three treatment replicates and one control per block were run to reduce experimental variability. Data collection and analysis were conducted in the same way as the larvae/temperature experiments and the egg/packaging material experiments, but the analysis was run in two separate parts (packaging and surface components) due to the differences in construction of treatment arenas.

Construction of treatment arenas for testing surface materials for wandering larvae required a different approach from that described in previous experiments. A method to contain the wandering larvae, and also to prevent escape from the treated surface, was needed; so that boxes were completely lined with the surface material. Wooden boxes were constructed with plywood. These were fastened together with wood adhesive and they had an inner surface area of

4 x 3.26 x 2.44 cm. The inside surfaces and lids of these wooden boxes were coated with the desired surface material. The self-adhesive vinyl floor tile was cut to size and pressed into the cubes. The concrete was mixed thick and pored into the boxes using a metal pan as a mold covered with plastic wrap. For the wood treatment, the boxes were lightly sanded. For the wood and tile surface treatments, arenas were caulked to smooth the box joints and to prevent escape from the treated surface. Because of the differences in surface types and box construction, boxes had slightly different inner dimensions for which the insecticide treatment was adjusted accordingly. Lids for the wooden boxes were a 5 x 5 cm piece of plywood; for the concrete treatment, a 5.75 x 4.5 cm poured concrete slab was used; and for the tile, a 5 x 5 cm piece of the vinyl flooring. Boxes were treated and larvae added in the same manner as above. Lids were sealed to the boxes using window caulk, which prevented escape but was easily removed after the exposure interval. To reduce variability, this experiment was run with two complete blocks with three treatment replicates and one control per block.

Results

Experiment 1: Survival of Eggs Exposed to Methoprene at Different Temperatures

There was a significant (P < 0.0002) effect of methoprene rate on Indianmeal moth survival. However, neither temperature nor the temperature by methoprene rate interaction was significant (Table 1-1). In general, there were very few significant pairwise differences in adult survival across treatments or replications except at 16°C where temperature was too low to support development, and at the highest rate of methoprene at 32°C where survival was lowest (Figure 1-1). (Mean separation analysis revealed that there was less than a 10% difference in survival between 20-28°C and 32°C.) Between 24 and 32°C mean adult emergence was significantly lower (P<0.05) at the 1.5x rate of methoprene compared to the corresponding lower rates or the untreated controls. This difference was not observed at 20°C (Figure 1-1).

Temperature had a significant effect on time to develop to adults; development was inversely related to temperature and survival to adulthood was significantly affected by temperature (P=0.0469) (Tables 1-2 and 1-3). The rate of methoprene did not influence time to first adult emergence (P=0.7763), but it did have a significant effect (P<0.005) on time until adults finished emerging. There was no significant temperature by treatment interaction (Tables

1-2 and 1-3). For all rates of methoprene, time to hatch decreased in a temperature-dependent manner between 20 and 28°C. However, hatching rates were similar at 28 and 32°C (Fig. 1-2).

Experiment 2: Survival of Fifth Instars Exposed to Methoprene at Different Temperatures

Fifth instars were sensitive to methoprene exposure and exposure interval (Table 1-4 and 1-5, P < 0.0001). There was a significant interaction between methoprene treatment and exposure interval (Table 1-4, 1-5). Between 20 and 32°C, survival to adult emergence was significantly lower when 5th instars were exposed to methoprene compared to the untreated control (Fig. 1-3). However, differences in survival among exposure intervals generally were not significant except at 32°C. The highest mortality occurred at the highest temperature and highest concentration similar (Fig. 1-3).

Figures 1-4, 1-5, and 1-6 display the results of Experiment B (separate graphs for 22, 27 and 32°, respectively). Table 5 is the analysis of variance that corresponds to these figures. All three main effects -- temperature, level of methoprene treatment, and exposure interval – significantly influenced survival to the adult stage. However, there were no significant 2- or 3-way interactions (Table 1-5). At all three temperatures, survival in both methoprene treatments was lower at the two longest exposure intervals than at the shortest or no exposure intervals (Figs. 1-4 to 1-6). Generally, differences occurred between the control and the highest application rate; but contrasts were only moderate between the control and the label rate.

Experiment 3: Survival of Eggs Exposed to Methoprene on Different Surfaces

Both main effects -- surface type/packaging materials and level of methoprene treatment (either the label rate or distilled water control) -- had a highly significant effect on survival (P<0.0009 and P<0.0001, respectively). There was also a highly significant interaction between these two factors (P<0.0001) (Table 1-6). Survival to adults was lowest when methoprene was applied to black paper with a laminate coating $(5.2 \pm 2.2\%)$ and to wood $(23.75 \pm 5.2\%)$ (Fig. 1-7). These two treatments also differed most from their respective controls. Survival was highest where methoprene was applied to plastic, tile, wax paper and uncoated cardboard (~60-63%). On most of these materials, survival was statistically no different than the untreated controls at P=0.05 (Fig. 1-7).

Experiment 4: Survival of Fifth Instars Exposed to Methoprene on Different Surfaces

Treatment of 5th instars with methoprene at the label rate (vs. no treatment) on different types of packaging material resulted in significant differences in survival (Table 1-7; methoprene: P<0.0001, packaging: P<0.0002). There was also a highly significant interaction between methoprene treatment and packaging type (Table 1-7). In contrast, exposure interval (2, 4 or 8 h at 27°C) had no influence on survival. However, there was a significant treatment by exposure interval interaction (Table 1-7). For all surfaces, differences between treatments and controls became larger with increasing exposure interval (cf. Figs. 1-8, 1-9, and 1-10). Adult emergence was consistently the lowest on laminated cardboard compared with its control across all exposure intervals (Figs. 1-8, 1-9, and 1-10). When concrete, wood and tile were tested, there was no general effect of surface material on survival of 5th instars through adult emergence (Table 1-8; P=0.202). However, methoprene treatment and exposure interval were significant (P < 0.0001 and P=0.0058, respectively). Of the three surface materials evaluated, lowest adult emergence occurred on the vinyl tile across all exposure intervals (Fig. 1-11).

Partial Budget Analysis

Using chemical cost information calculated per 929 m² (10,000 ft²) for surface application, we conducted a partial budget analysis to compare costs of methoprene as a surface treatment for eggs and larvae at several rates and exposures. Economic risk was calculated at three levels (90, 95, and 99%) as deviations below the target goal. In the case of these three experiments, time and equipment cost were fixed and the only variable cost was the cost of the chemical. Risk, in this case, was the inverse of mortality up to the mortality target, at which it was set to zero. Therefore, the downside risk was mortality below the target level. Results are displayed in Table 1-9. Results show that for eggs, using the no action (untreated control) strategy was virtually as effective as either of the realistic application rates (0.5x and the label rate). For wandering larvae, methoprene was more effective; with increasing exposure interval, risk decreased but total chemical cost remained the same.

Discussion

Overall, temperature does not affect the efficacy of methoprene as a surface treatment for control of the Indianmeal moth. This finding is similar to that of hydroprene and temperature on

survival of Indianmeal moth to the adult stage (Mohandas et al, 2006b). The poor control of eggs and wandering larvae of Indianmeal moth is in contrast to the work of Arthur (2004) and Chanbang et al. (2007) where methoprene was highly effective on progeny of adults of *Rhyzopertha dominica* exposed on grain. It is also in contrast to the work conducted by Mohandas et al. (2006b, 2006c) where hydroprene (another juvenile hormone mimic) was used with success to control eggs and wandering-phase larvae of the Indianmeal moth. Hydroprene has also been shown effective against larval *Tribolium castaneum* (Arthur 2001, Toews et al. 2005).

For eggs of the Indianmeal moth, methoprene is most likely not a reliable control strategy, as overall survival to the adult stage was high (~78 to 88% in all treatments compared to ~88% for the controls). Fifth instars appear to be a more susceptible life stage to methoprene than eggs. However, survival for treated 5th instars (~54%) was still quite high compared to the controls (~90%). In fact, given the low tolerance for Indianmeal moths and stored product pests in general, this level of adult survival would be considered unacceptable by industry standards. Survival to adult was reduced when larvae were exposed for two hours or more at temperatures ranging from 22-32°C; but survival was still 51%. My findings suggest that mortality of 5th instars may be increased with repeated or prolonged exposure; but more data are needed.

Although manufacturers may not be directly responsible for the damage done to packaging in shipment, they remain accountable for the finished products (Highland 1978). Economic losses as a result of insect infestation in finished products exceed the loss of quantity and quality that is easily quantified, but can extend to loss of consumer confidence which, ultimately, may tarnish the company's image in the eyes of consumers. The need for protection of finished stored products in storage and transit is essential. There are many ways to decrease the incidence of insects entering finished product packages, including the use of repellents, odor barriers, insecticides and insect-resistant packaging (Highland 1978, Mullen 2000, Mullen and Mowery 2003). Surface and package material treatment with methoprene or similar IGRs may be an effective control strategy for other insects such as flour beetles (Arthur, 2004). But based on my studies, it appears to provide only marginal protection against the Indianmeal moth. While the use of methoprene on finished product packaging materials may not prevent entry into packages, it could partially suppress infestations by preventing subsequent generations from infesting other packages in the shipment or warehouse.

There may be several reasons for the high survival of Indianmeal moth eggs and larvae on treated package materials. Methoprene is considered to be relatively stable over a long period of time in grain bins and over a wide range of temperatures (Daglish et al. 1995, Daglish and Wallbank 2005); so degradation of the insecticide most likely is not related to the relatively high survival I observed. On the other hand, differences in surface materials may be an important factor. Absorption of methoprene into the package material may not leave enough active ingredient on the surface for neonates and wandering larvae to acquire. This may correspond to the lower survival of Indianmeal moths on non-porous materials such as vinyl flooring tile and laminated paper package treatments compared with porous materials such as wood and uncoated Kraft paper. Lower survival to the adult stage in the surface treatments with eggs may also be attributed to the application method (airbrush versus dispersing with a glass rod); there may have been better coverage with the airbrush treatment method.

Other studies (Jenson, unpublished data) show that aerosol applications of methoprene reduce survival to the adult stage in eggs of the Indianmeal moth exposed directly to the insecticide and on package materials exposed to the insecticide. Aerosol applications of methoprene are formulated with an organic solvent and this may partially explain the differences with this study where methoprene is formulated with water for surface treatments. Aerosol treatments of methoprene have also recently been shown to be effective against flour beetles (Arthur 2008). While methoprene as a surface treatment is not likely to replace aerosol insecticides or fumigants for control of this pest, it could be a useful part of an overall integrated pest management program.

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Figures and Tables

Figure 1-1 Survival to the adult stage of Indianmeal moth eggs exposed to four rates of methoprene (0 (control), the label rate for surface application (1 ml/ $94m^2$), 0.5 times the label rate, and 1.5 times the label rate) and held under five constant temperatures 16, 20, 24, 28, and 32°C (16 °C data not shown). Asterisks indicate treatments that were statistically different from other treatments at P= 0.05. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P =0.05 for each treatment combination.

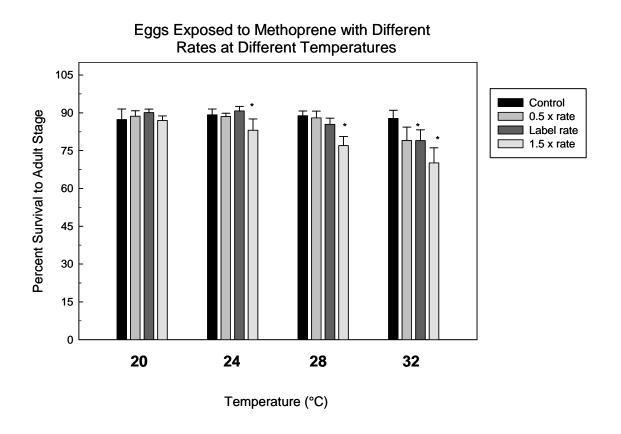


Figure 1-2 Days until 75% hatch, beginning and ending adult emergence on Indianmeal moth eggs exposed to four rates of methoprene (0 (control), the label rate for surface application (1 ml/ 94m²), 0.5 times the label rate, and 1.5 times the label rate) and held under five constant temperatures 16, 20, 24, 28, and 32°C (16°C data not shown). Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P =0.05 for each treatment combination. Capital letters indicate statistical differences in the temperature treatment, there was no significant difference at P=0.05 for insecticide treatments.

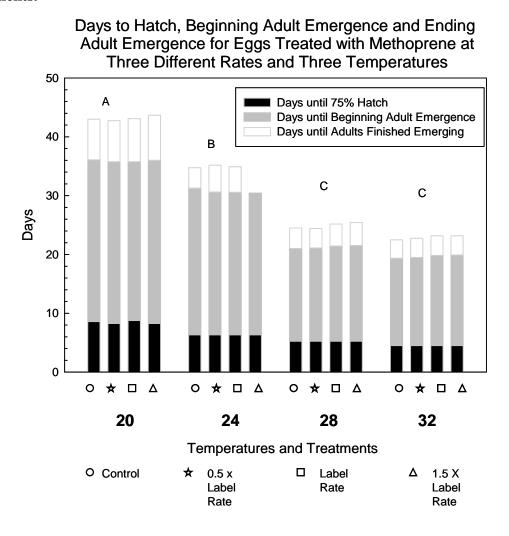
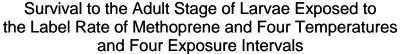
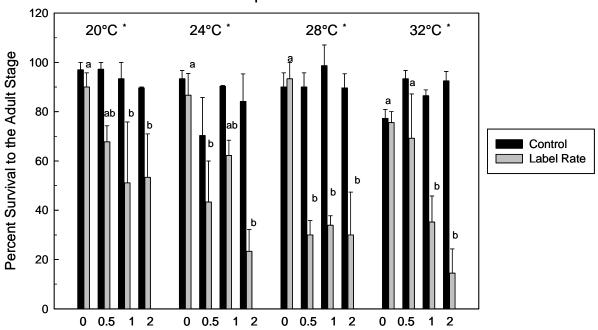


Figure 1-3 Survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface application at four temperatures (20, 24, 28, and 32° C) and four exposure intervals (0, 0.5, 1, and 2 hours). Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Lower case letters indicate statistical differences among treatment*temperature combinations at each exposure interval. There was no statistical differences in survival of controls at any temperature or exposure interval P = 0.05 level.





Exposure Intervals (in hours) by Temperature

Figure 1-4 Survival to the adult stage of 5^{th} instars exposed to four concentrations (0.5x the label rate, the label rate, 1.5x the label rate, and a control) of methoprene at five exposure intervals (0, 0.5, 1, 2, and 4) and held at 22° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference among controls across treatments and exposure intervals and lower case letters indicate differences by level of insecticide treatment across exposure intervals. Treatments were statistically different from controls but not from each other at 2 and 4 hour exposure intervals.

Larvae Exposed to Three Rates of Methoprene at Five Exposure Intervals at 22°C

Exposure Interval

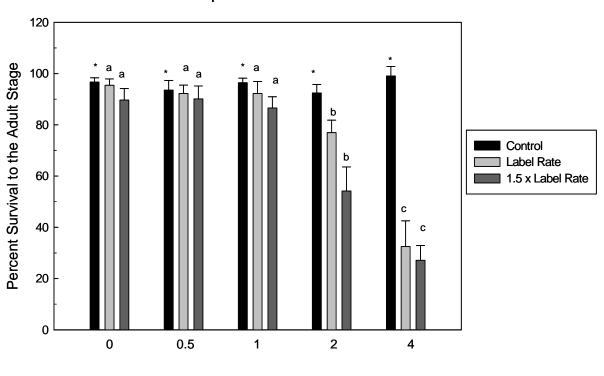
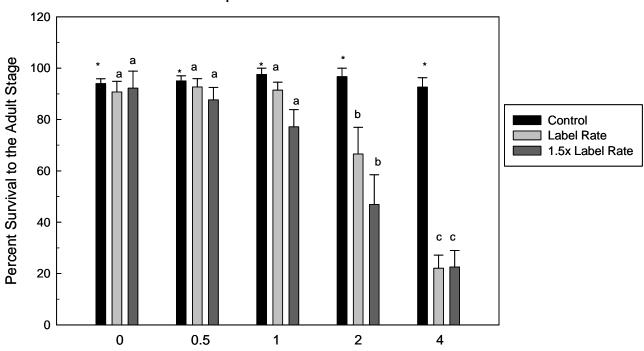


Figure 1-5 Survival to the adult stage of 5^{th} instars exposed to four concentrations (0.5x the label rate, the label rate, 1.5x the label rate, and a control) of methoprene at five exposure intervals (0, 0.5, 1, 2, and 4) and held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference among controls across treatments and exposure intervals and lower case letters indicate differences by level of insecticide treatment across exposure intervals. Treatments were statistically different from controls but not from each other at 2 and 4 hour exposure intervals.

Larvae Exposed to Three Rates of Methoprene at Five Exposure Intervals at 27°C



Exposure Interval

Figure 1-6 Survival to the adult stage of 5^{th} instars exposed to four concentrations (0.5x the label rate, the label rate, 1.5x the label rate, and a control) of methoprene at five exposure intervals (0, 0.5, 1, 2, and 4) and held at 32° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference among controls across treatments and exposure intervals and lower case letters indicate differences by level of insecticide treatment across exposure intervals. Treatments were statistically different from controls but not from each other at 2 and 4 hour exposure intervals.

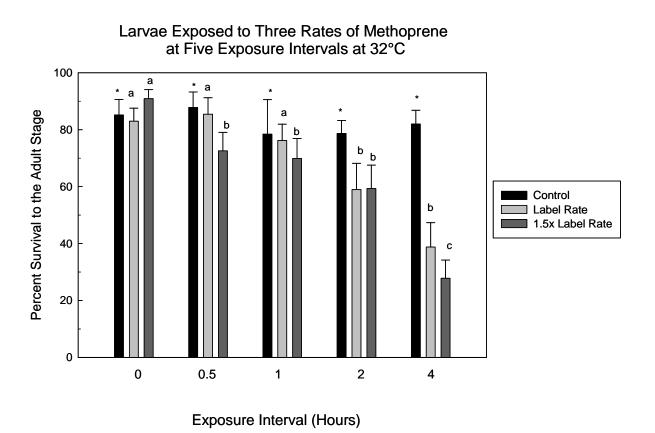
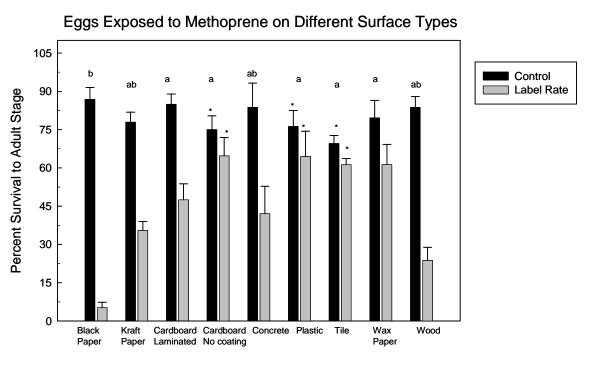


Figure 1-7 Eggs exposed to the label rate of methoprene for surface applications on nine different packaging and surface types held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Lower case letters indicate statistical differences between surface type treatment and asterisks indicate treatment pairs where there were not statistical differences in the between treatments and controls at the P = 0.05 level.



Surface and Packaging Materials

Figure 1-8 Survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface applications on six different packaging types on a 2-hour exposure interval, held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference between treatment and controls and lower case letters indicate differences in survival when exposed to the label rate of methoprene with packaging type.

Two Hour Exposure to Methoprene at the Label Rate by Surface Type

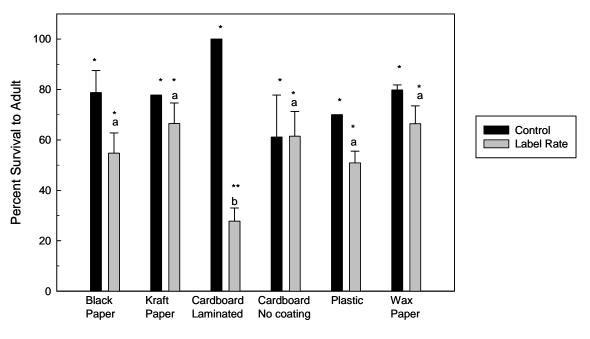
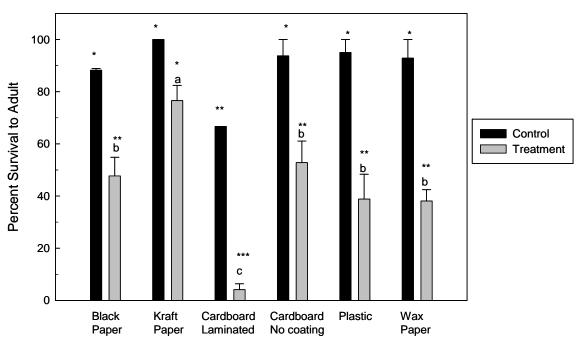


Figure 1-9 Survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface applications on six different packaging types on a 4-hour exposure interval, held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference between treatment and controls and lower case letters indicate differences in survival when exposed to the label rate of methoprene with packaging type.

Four Hour Exposure to Methoprene at the Label Rate by Surface Type



Surface Type

Figure 1-10 Survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface applications on six different packaging types on an 8-hour exposure interval, held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference between treatment and controls and lower case letters indicate differences in survival when exposed to the label rate of methoprene with packaging type.

Eight Hour Exposure to Methoprene at the Label Rate by Surface Type

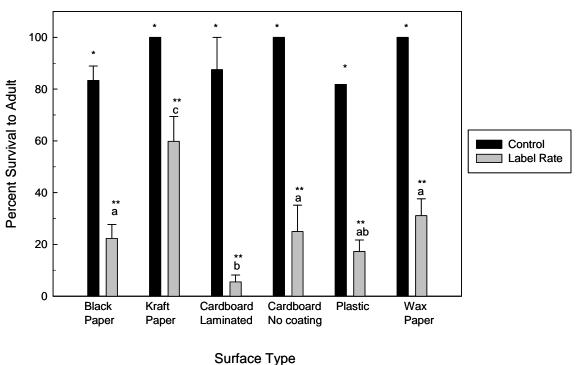


Figure 1-11 Survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface applications on three different surface types on three exposure intervals (2, 4, and 8 hours) held at 27° C. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Asterisks indicate statistical difference between treatment and controls across all surface types and exposure intervals.

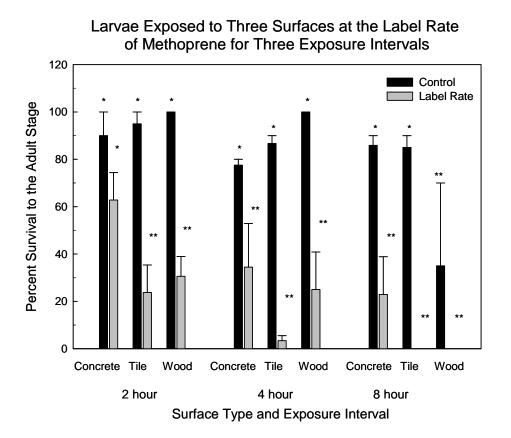


Table 1-1 Results of analysis of variance on survival of Indianmeal moth eggs exposed to four rates of methoprene (0 (control), the label rate for surface application (1 ml/ 94m²), 0.5 times the label rate, and 1.5 times the label rate) and held under five constant temperatures 16, 20, 24, 28, and 32°C (16°C not included in analysis due to extremely low survival).

Effect of Methoprene and Temperature	Num DF	Den DF	F Value	Pr > F
Temperature	3	4.91	1.39	0.3501
Level of Methoprene Treatment	3	150	7.03	0.0002
Temperature*Treatment	9	150	0.94	0.4946

Table 1-2 Results of analysis of variance on days to beginning adult emergence on Indianmeal moth eggs exposed to four rates of methoprene (0 (control), the label rate for surface application (1 ml/ $94m^2$), 0.5 times the label rate, and 1.5 times the label rate) and held under five constant temperatures 16, 20, 24, 28, and 32° C (16° C not included in analysis due to extremely low survival).

Effect on Days to First Adult Emergence	Num DF	Den DF	F Value	Pr > F
Temperature	3	6	4.91	0.0469
Level of Methoprene Treatment	3	167	0.37	0.7763
Temperature*Treatment	9	167	0.75	0.6634

Table 1-3 Results of analysis of variance on days to final adult emergence on Indianmeal moth eggs exposed to four rates of methoprene (0 (control), the label rate for surface application (1 ml/ $94m^2$), 0.5 times the label rate, and 1.5 times the label rate) and held under five constant temperatures 16, 20, 24, 28, and 32° C (16° C not included in analysis due to extremely low survival).

Effect on Days until Ending Adult	Num DF	Den DF	F Value	Pr > F
Emergence				
Temperature	3	6	10.03	0.0094
Level of Methoprene Treatment	3	167	4.38	0.0054
Temperature*Treatment	9	167	0.42	0.9250

Table 1-4 Results of analysis of variance on survival to the adult stage of 5^{th} instars exposed to the label rate of methoprene for surface application at four temperatures (20, 24, 28, and 32° C) and four exposure intervals (0, 0.5, 1, and 2 hours).

Effect	Num DF	Den DF	F Value	Pr > F
Temperature	3	64	0.00	1.0000
Level of Methoprene Treatment	1	64	107.10	<.0001
Temperature * Treatment	3	64	1.21	0.3117
Exposure Interval	3	64	11.61	<.0001
Temperature * Exposure Interval	9	64	1.66	0.1171
Treatment * Exposure Interval	3	64	11.92	<.0001
Temp. * Treatment * Exposure Interval	9	64	1.04	0.4167

Table 1-5 Results of analysis of variance on survival to the adult stage of 5^{th} instars exposed to four concentrations (0.5x the label rate, the label rate, 1.5x the label rate, and a control) of methoprene at five exposure intervals (0, 0.5, 1, 2, and 4) and held at three temperatures 22, 27 and 32°C. Corresponds with Figures 1-5 to 1-7.

Effect	Num DF	Den DF	F Value	Pr > F
Temperature	2	4	9.06	0.0327
Level of Methoprene Treatment	2	354	72.32	<.0001
Temperature * Treatment	4	354	1.40	0.2340
Exposure Interval	4	354	79.50	<.0001
Temperature * Exposure Interval	8	354	1.11	0.3581
Treatment * Exposure Interval	8	354	19.62	<.0001
Temp. * Treatment * Exposure Interval	16	354	0.90	0.5695

Table 1-6 Results of analysis of variance on eggs exposed to the label rate of methoprene for surface applications on nine different packaging and surface types held at 27° C.

Effect	Num DF	Den DF	F Value	Pr > F
Surface or Packaging Materials	8	51	4.05	0.0009
Level of Treatment	1	51	150.32	<.0001
Surface Material * Treatment	8	51	8.78	<.0001

Table 1-7 Results of analysis of variance on survival to the adult stage of $5^{\rm th}$ instars exposed to the label rate of methoprene for surface applications on six different packaging types (Kraft paper, black paper, waxy paper, cardboard with laminated coating, cardboard with no coating, and plastic) on three exposure intervals (2, 4, and 8 hours) held at 27° C. Corresponds with Figures 1-8 to 1-10.

Effect	Num DF	Den DF	F Value	Pr > F
Level of Methoprene Treatment	1	177	137.92	<.0001
Exposure Interval	2	177	0.16	0.8546
Treatment * Exposure Interval	2	177	12.55	<.0001
Package Material	5	177	5.20	0.0002
Treatment * Package Material	5	177	2.56	0.0292
Exposure Interval * Package Material	10	177	1.61	0.1060
Treatment * Exposure Interval* Package Material	10	177	1.62	0.1031

Table 1-8 Results of analysis of variance on survival to the adult stage of 5th instars exposed to the label rate of methoprene for surface applications on three different surface types (concrete, wood and tile) on three exposure intervals (2, 4, and 8 hours) held at 27°C.

Effect	Num DF	Den DF	F Value	Pr > F
Level of Methoprene Treatment	1	53	76.22	<.0001
Exposure Interval	2	53	5.69	0.0058
Treatment * Exposure Interval	2	53	0.21	0.8119
Surface Material	2	53	1.65	0.2021
Treatment * Surface	2	53	2.13	0.1293
Exposure Interval * Surface	4	53	1.46	0.2284
Treatment * Exposure Interval* Surface	4	53	0.85	0.4999

Table 1-9 Summary of costs and risk levels for methoprene surface treatments compared with untreated controls for Experiments 1 and 2a. Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Letters denote statistical difference at the P= 0.05 level. The mean survival of the egg stage are separated using insecticide treatment (Experiment 1) and larvae by insecticide treatment and exposure interval, since temperature wasn't statistically significant for survival in Experiments 1 and 2a. The scenario that represents the lowest risk, lowest cost and highest mortality are the highest levels of methoprene and the longest exposure intervals.

Leonon	nic					
Summa						
Costs: p	er $10,000$ ft ² (label rate)	Methoprene = \$1	.56			
F 11		CM d				
-	Eggs, Temperature and Rat Treatment			Dial. (050/)	Digl. (000/.)	Cost (\$)
Stage		% Mortality	Risk (90%)	Risk (95%)	Risk (99%)	<i>Cost</i> (\$)
Eggs	Untreated Control	0.116896 ^A	0.783104	0.833104	0.873104	0.00
Eggs	0.5x Rate	0.122561 ^A	0.777439	0.827439	0.867439	0.78
Eggs	Label Rate	0.156433 ^A	0.743567	0.793567	0.833567	1.56
Eggs	1.5x Rate	0.218588^{B}	0.681412	0.731412	0.771412	2.34
Lggs			0.001.12	****	*** * * * * * * * * * * * * * * * * * *	

Exp. 2	Larvae, Temperature, Rate	e of Methoprene,	and Exposure Interv	val		
	Larvae, Temperature, Rate Treatment	e of Methoprene,			Risk (99%)	Cost (\$)
Exp. 2 Stage	Treatment	e of Methoprene, % Mortality	and Exposure Interv	val		
Exp. 2	Treatment Untreated Control at all	e of Methoprene, % Mortality	and Exposure Interv	val		
Exp. 2 Stage	Treatment	e of Methoprene, % Mortality	and Exposure Interv Risk (90%)	Risk (95%)	Risk (99%)	Cost (\$)
Exp. 2 Stage	Treatment Untreated Control at all exposures	e of Methoprene, % Mortality 0.104346 ^A	and Exposure Interv Risk (90%) 0.795654	Risk (95%) 0.845654	Risk (99%) 0.885654	Cost (\$)
Exp. 2 Stage Larvae Larvae	Treatment Untreated Control at all exposures Label Rate * 0 hr	e of Methoprene, % Mortality 0.104346 ^A 0.136111 ^A	and Exposure Interv Risk (90%) 0.795654 0.763889	Risk (95%) 0.845654 0.813889	Risk (99%) 0.885654 0.853889	Cost (\$) 0.00 1.56

CHAPTER 2 - Efficacy of an Esfenvalerate plus Methoprene Aerosol for the Control of Eggs and Fifth Instars of the Indianmeal moth (Lepidoptera: Pyralidae)

Abstract

To fulfill the mandate to replace the fumigant methyl bromide, new options for managing stored product pests in food manufacturing and storage facilities are needed. Aerosol insecticides provide one alternative, but little is known about the efficacy of these insecticides for control of the Indianmeal moth, *Plodia interpunctella* (Hübner), a major insect pest of stored processed food. One insecticide combination currently in use commercially to control stored-product insects is the pyrethroid, esfenvalerate, in combination with the insect growth regulator (IGR), methoprene. Pyrethroids are contact insecticides that kill adult stored-product insects, but aerosol formulations may not provide residual efficacy. Methoprene is an IGR that normally does not affect adults, but has residual persistence for control of immature stages of most storedproduct insects when applied as a liquid contact spray or as an aerosol formulation. In this study, eggs and 5th instars of the Indianmeal moth were exposed to aerosol applications of esfenvalerate and methoprene, alone and in combination, in open and obstructed positions inside small sheds. When larvae were exposed to methoprene, subsequent adult emergence was $7.1 \pm 1.5\%$. In contrast, adult emergence was $92.5 \pm 3.5\%$ when larvae were treated with esfenvalerate alone. When eggs were exposed to methoprene, subsequent adult emergence ranged from $10.8 \pm 1.3\%$ to $44.8 \pm 14.3\%$; but when eggs were exposed to esfenvalerate, adult emergence was consistently low (13.8 \pm 10.8%). This emergence rate also was considerably lower than when fifth instars were exposed to esfenvalerate. In the combination treatment of methoprene plus esfenvalerate at their respective label rates, adult emergence following larval exposure was $0.91 \pm 0.61\%$ compared to $16.3 \pm 9.6\%$ when eggs were exposed. Our results indicate that while methoprene

alone is highly effective in reducing adult emergence after larval exposure, it is not as effective on eggs as esfenvalerate. Results suggest that different combinations of pyrethroid plus methoprene at less than the label rate could be used to control both egg and wandering-phase larval stages of the Indianmeal moth. An economic risk analysis also supports a strategy of combining methoprene and esfenvalerate

Keywords: methoprene, esfenvalerate, aerosols, Indianmeal moth

Introduction

New strategies for managing stored product pests are needed to replace the fumigant methyl bromide in food manufacturing and storage facilities, especially for control of the Indianmeal moth, *Plodia interpunctella* Hübner. These pyralid moths can be problematic in all life stages and in all phases of the food manufacturing process, from grain bin to end user (Doud and Phillips 2000, Johnson et al. 2003, Mahroof and Subramanyam 2006). Indianmeal moth infestations of finished stored products present a unique challenge in that the products typically are of high-value products and are stored for variable periods of time in multiple locations. Thus, the insect threshold is essentially zero. The Indianmeal moth is a cosmopolitan pest known to infest a great number of commodities, including many different grains, dried fruit and nuts (Mohandass et al. 2007). Infestations can lead to equipment damage, physical product losses, aesthetic damage and unpleasant odors. Even one insect can cause negative consumer feedback, which compounds problems related to the food manufacturing process.

We have identified two main windows of time in the Indianmeal moth life cycle for controlling them in finished stored products with aerosol insecticides. Unlike fumigants, aerosols do not infiltrate packaging materials; so treatment needs to be made when the Indianmeal moth is on the exterior of a package. The first window for control is late instar wandering-phase larvae as they leave the food source to find a pupation site. The second is eggs that are oviposited in unconcealed areas that may come into contact with a surface or aerosol application of insecticide.

The Indianmeal moth is generally difficult to control with conventional insecticides, especially in the late-larval stages (Arthur 1997). For example, Arthur (1997) showed that 5th instars are insensitive to residues of deltamethrin dust except during the immediate period after treatment. Moreover, cyfluthrin and chlorpyrifos-methyl are only effective at high rates (22-30 ppm) and immediately following application (Arthur 1989, 1995).

Eggs of stored grain and fruit pests are often the most difficult life stage to kill using conventional fumigants, including methyl bromide (Weller and Morton 2001, Armstrong and Whitehand 2005). Eggs of the Indianmeal moth are also an important target life stage for control as they are relatively heat- and cold-tolerant, making them difficult to control with non-chemical

management strategies (Mahroof and Subramanyam 2006, Johnson 2007). With high rates of application, low residual activity of insecticides, and the apparent insensitivity of 5th instars to conventional insecticides, a replacement for fumigants is needed to control the Indianmeal moth in areas where processed and packaged foods are stored. One insecticide combination currently in use is the pyrethroid, esfenvalerate, in conjunction with methoprene. Pyrethroids work as contact insecticides whereas methoprene provides residual control.

One constraint on integrated pest managers is cost. Chemicals, applicator labor and/or equipment costs, as well as costs associated with shutdown/loss of production in terms of time, are a major consideration. Partial budget analysis compares costs of replacement control strategies with existing ones. Thus, they are used to determine levels of economic risk associated with each strategy (Boehlje 1984). A partial budget analysis can compare different application methods and rates of methoprene alone and in combination with conventional insecticides such as esfenvalerate to enable food production plant managers and warehouse managers to make better decisions. Economic analysis of other methyl bromide alternatives using enterprise budgets for specific field crops have been developed (Nelson 1996, Byrd et al. 2006).

The objectives of this study were to 1) determine the effect of aerosol application of methoprene alone and in combination with an esfenvalerate insecticide on the survival of Indianmeal moth to the adult stage, and 2) estimate differences in cost and risk associated with each strategy.

Materials and Methods

General Procedures

The Indianmeal moths used in the experiments derive from a laboratory colony established in 1988 and periodically supplemented with wild individuals from collections in Riley County, Kansas in the United States. The colony is located at the USDA-ARS Grain Marketing and Production Research Center in Manhattan, KS¹ and has been maintained

¹ Kansas State University Museum of Entomological and Prairie Arthropod Research under Lot Number 208.

continually in environmental growth chambers (Forma-Scientific, Thermo Electron Corporation, Waltham, MA) at 27±1°C, approximately 40% RH., and in darkness (L:D = 0:24 hrs). Relative humidity was maintained using pans of water in the bottom of the incubator. Larvae have been reared on a standard enriched wheat diet consisting of cracked wheat and shorts (4.4 kg), brewer's yeast (22g), sorbic and benzoic acid (9.5g each), honey (240ml), glycerin (240ml) and water (120ml).

During experiments insects were placed on clean diet following treatment and allowed to develop and/or mature to the adult stage under the above environmental conditions. Three separate experiments were conducted using small sheds measuring 2.8 m wide x 5.9 m long x 2.0-2.2 m high to simulate a food warehouse situation. Flooring and drywall in each shed were installed in the same manner described by Toews et al. (2005a). Temperature and relative humidity were recorded using a HOBO data logger (Onset Computer Corporation, Bourne, MA) placed next to treatment areas.

In each shed, concealed conditions such as those found in food storage warehouse were created using large corrugated cardboard boxes placed on top of a wooden pallets suspended on concrete blocks. Concrete blocks were used to raise the pallets so that treatment arenas could be placed underneath the pallets. Eggs (less than 24 hours after oviposition) were exposed to the aerosol spray in 100 mm Petri dishes painted with acrylic paint to reduce static. Actively wandering 5th instars were exposed using a series (five sizes) of cardboard garment boxes nestled together and secured with a mixture of instant tapioca and water, to hold the boxes together. Boxes were then taped with painter's tape around the edges so that larvae could not escape the aerosol by crawling under the boxes. Box dimensions ranged from 21.94 cm - 53.34 cm (11-21 in) long and 21.59 -35.56 cm (8.5-14 in) wide; when stacked inside each other, the distance from each side was approximately 2.54 cm (1 in) from the next closest box edge. The glue mixture was previously determined to have no adverse affect on the larvae. These "hurdles" were designed to contain the wandering larvae during the two-hour exposure period (see Figure 2-1). Preliminary experiments were conducted to determine the viability of this treatment method.

Within each shed there were both unobstructed and concealed positions. The standard experimental set-up of each shed is illustrated in Figure 2-2. Unobstructed positions were considered to be those that were fully exposed to the settling aerosol particles, and concealed positions were those underneath the mock pallet, partially shielded from the settling particles.

Experiment 1

Applications were done using a hand-held ultra low volume (ULV) applicator (model no. E2 MLD® Chemical Dispersal Unit, MicroGen Equipment Corporation, San Antonio, TX 78217). Each of four sheds was treated with methoprene aerosol in proportion to the label rate (Central Sciences 2004) of 3 ml of the Diacon II® formulation, which is 900 mg of active ingredient [AI], per 283.7 m³ (10,000 ft³). The insecticide was formulated by mixing 1.93 ml methoprene with 200 ml of a petroleum-based carrier (Isopar-M). The target volume for application in the smaller three sheds was 35.7 ml diluted solution and 44.0 ml for the larger two sheds. Solutions were weighed prior to application and after to be sure that the target amount of chemical was applied to each shed. This corresponds with the label rate for aerosol applications, and the equipment dispenses aerosol at the rate of 29.5 ml per 60 s. One shed was chosen as the untreated control and the same shed was designated as the untreated control for both blocks of this experiment. The insecticide was applied by running the applicator for the time calculated to achieve the label rate for the two shed sizes (71 and 81 s). The applicator slowly pivoted during the spray to ensure even coverage. Timing was done by the applicator and also a person outside the treatment area.

The first block of this experiment was conducted in June of 2006 and the second completed in September 2006. Eggs and fifth instars were exposed to the aerosol in unobstructed and concealed positions with three replicates of each. In this experiment, twenty eggs were exposed using 100 mm Petri dishes lined with filter paper. Insects were placed in the treatment areas immediately prior to insecticide application. After the aerosol application was completed, the insects remained in place for two hours to be consistent with similar field applications of aerosols. The lights were turned off inside the sheds while the insects were exposed. Temperature and relative humidity were monitored during the two hour exposure period and temperatures were $23 \pm 1^{\circ}$ C with 60 ± 8 % relative humidity for the first block and $21\pm 1^{\circ}$ C with 60 ± 2 % relative humidity. After this two-hour exposure period, egg dishes and larvae were collected from arenas and transported back to the laboratory. Eggs were counted for each treatment and both eggs and larvae transferred to clean dishes with wheat diet and incubated at $27\pm 1^{\circ}$ C at approximately 40% relative humidity until they emerged as adults or

were determined to be unable to emerge. This determination was made when a second generation of larvae was visible in the control dishes (approximately one week after adults began to emerge in controls). Counts of emerged adults were made at three weeks, and a second count made at four weeks from date of exposure.

This experiment was designed as a split plot with the methoprene or untreated control treatment as the whole plot treatment and unobstructed and concealed positions as the sub-plot treatments. Data analysis was conducted using the Mixed Procedure (PROC Mixed) of the Statistical Analysis System (SAS) (SAS institute 2001). Means and standard errors for percent survival for each life stage were also calculated using the MEANS Procedure of SAS. Means for treatments were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P =0.05 for each for each life stage.

Experiment 2

In the second experiment, methoprene was compared to the pyrethroid esfenvalerate (Conquer®, (EPA Reg. No. 1021-1641-57076) and a combination treatment of esfenvalerate with methoprene. The esfenvalerate solution, applied in proportion to the label rate of 29.6 ml diluted solution per 28.3 m³ (1000 ft³), was made from 296 ml Conquer® concentrate in 3785.4 ml (1 gallon) carrier oil. This chemical was applied in the same manner and formulated as 15.47 ml esfenvalerate in 200 ml of oil for applying to the sheds. The target delivery rate for esfenvalerate alone was similar to the methoprene alone treatments. This experiment was conducted in the same manner as Experiment 1; however, approximately 50 eggs were exposed to the aerosol in painted 100 ml Petri dishes. Following treatment, 20 eggs were selected from each treatment and allowed to mature to the adult stage. Six blocks by time replicates were conducted, with only one replication of each life stage at each position. Four sheds were assigned to the chemical treatments: 1) carrier only, 2) methoprene at the label rate, 3) esfenvalerate at the label rate, and 4) a combination of methoprene and esfenvalerate at their respective label rates. In addition, one shed was designated as the untreated control. Although we had the ability to regulate temperature within each shed, air conditioners remained turned off so that air flow would not interfere with the aerosol treatment. Blocks were conducted in late May 2007 through early July 2007 with temperatures and relative humidity recorded by HOBO monitors. Temperatures ranged from 21 to 27 + 1°C and relative humidity ranged from 40-60%.

This variation in the physical environment was due to the varying times of the day and the period of time over which the tests were conducted. In this experiment, treatments were fixed to each shed to avoid contamination between sheds. Any larvae that escaped the treatment arenas were disposed of and not included in the analysis. Data analysis was conducted as in Experiment 1.

Experiment 3

In this experiment, different combinations of methoprene and esfenvalerate at less than the label rate were assessed to determine if treatments at less than the label rate could be as effective full rates. Four chemical treatments: 1) full label rate methoprene plus one-third of the label rate esfenvalerate, 2) full label rate methoprene plus two-thirds of the label rate esfenvalerate, 3) full label rate esfenvalerate plus one-third of the label rate methoprene, and 4) full label rate esfenvalerate plus two-thirds of the label rate of methoprene, were conducted and, again, one shed served as the untreated control. Treatments were assigned to each shed in a Latin square blocked by day of treatment. We conducted each block during a different time during the day with block one run in September 2007 and blocks 2-5 in September 2007. Temperatures were monitored using HOBO data monitors. Temperatures ranged from 20 to 27 ± 1°C and relative humidity ranged from 45 to 60%. Five complete blocks were conducted with one replicate of each life stage by position sub-plot treatment. The experimental design was split plot and data were analyzed using Proc Mixed in SAS.

Results

There were no significant differences in adult emergence for either exposed eggs or 5^{th} instars between the concealed and unobstructed (open) habitats in any of the experiments (Tables 2-1, 2-3). In the methoprene-only treatments, survival to the adult stage was significantly lower (P < 0.0001) for both eggs and 5^{th} instars compared to the untreated controls (Fig. 2-2). For exposed eggs, $10.8 \pm 9.1\%$ emerged compared to $72.1 \pm 9.9\%$ in the control; for fifth instars, the emergence rate was $7.1 \pm 10.6\%$ compared to $87.7 \pm 12.5\%$ in the control. Overall, adult emergence was significantly different (P = 0.0149) in regard to eggs versus larvae.

When Indianmeal eggs were exposed to each insecticide alone, or a combination of the two, adult emergence was significantly lower (mean range: \sim 18-30%) than in the untreated control (83.7 + 5.8%) or with carrier (67.3 + 32.4%) (Fig. 2-3). Emergence also was

significantly lower when exposed to the carrier than to the untreated control. There was no statistical difference in adult emergence between the methoprene and esfenvalerate treatments (both \sim 38%); but these chemicals in combination reduced adult emergence to approximately half of that in either chemical treatment alone (Fig. 2.3).

There was a large difference in larval response to insecticide treatments; fifth instars that were exposed to methoprene alone had a very low emergence rate $(0.4 \pm 1.4\%)$. However, adult emergence in the esfenvalerate treatment was $90.4 \pm 9.3\%$, which was not statistically different from the untreated control (P = 0.7055) (Fig. 2.3). When methoprene was combined with esfenvalerate, emergence was very low $(1.98 \pm 2.35\%)$ and statistically similar to the methoprene treatment (mean $0.417 \pm 1.44\%$). The carrier did not reduce adult emergence compared to the untreated control after 5th instar exposure $(87.8 \pm 10.1$ and $89.5 \pm 10.9\%$, respectively) (Fig. 2.3).

All rates and combinations of insecticides resulted in reduced survival of Indianmeal moths compared to the untreated control, both when eggs and 5th instars were exposed (P<0.0001). However, for each life stage (eggs or 5th instar), no combination of treatments had a significantly different effect on survival ($P \le 0.05$). While high mortality is the measure of efficacy of any insecticide, cost is also a major consideration in any pest management program. Using chemical cost information calculated per 283.7 m³ (10,000 ft³), we conducted a partial budget analysis to compare costs of methoprene and esfenvalerate treatments. Economic risk was calculated at three levels (90, 95, and 99%) as deviations below the target goal. Tilley (2007) reports a modified Target MOTAD (mortality goal) model for optimizing cost and risk, but in the case of these three experiments, time and equipment cost are fixed and the only variable cost is the cost of the chemical. Risk, in this case, is the inverse of mortality up to the mortality target, at which it is set to zero. Therefore, the downside risk is mortality below the target level. Results are displayed in Tables 2-4 through 2-6. Chemical and carrier oil costs were calculated based on current industry prices. Carrier oil costs fluctuate with the global petroleum market but for the purposes of this analysis were fixed to \$0.83 per L (\$3.15 per gallon) or \$0.0008 per ml. Our results indicate that while methoprene alone reduced adult emergence in treated larvae by 99.58 %, it only reduced emergence for treated eggs by 26.33%. Conversely, the esfenvalerate treatment reduced adult survival by 9.67% for exposed larvae, and reduced emergence in exposed eggs by 64.58%. Of all treatment scenarios, the full rate methoprene plus full rate esfenvalerate treatments represent the lowest risk and also represent the highest cost per 283.7 m³ (10,000 ft³) for controlling both life stages of the Indianmeal moth (Table 2-6).

Discussion

Overall, aerosol methoprene and esfenvalerate treatments had good efficacy, both alone and in combination, for eggs and fifth instars of the Indianmeal moth. Adult emergence was significantly different in regard to eggs versus larvae, which is not surprising given the hardiness of wandering phase Lepidoptera larvae and the need to transport insects to the research site and back to the laboratory. These results show that eggs have slightly higher survival to the adult stage in the combination treatments (experiment 3) than in experiment 2; however there were no treatments in experiment 3 that contained the full rates of both methoprene and esfenvalerate. It is interesting to note that the life stages (egg and fifth instar) responded very differently to the methoprene and esfenvalerate treatments. It could reasonably be concluded that for overall control of the Indianmeal moth, the use of both chemicals together would result in greater control of both life stages than either chemical alone.

These results add to other recent publications demonstrating the efficacy of aerosol treatments (Arthur and Campbell 2007, Arthur 2008) for both conventional insecticides and insect growth regulators. The other significant result is that there was no difference between percent mortality in unobstructed and concealed positions for any treatment combination. This is consistent with other studies (Arthur and Campbell 2007, Arthur 2008) showing good coverage of the aerosol fog at least as far as 0.3048 m (1 ft) under equipment and pallets. This study simulates some possible field conditions, as food manufacturing and storage facilities often have product stacked on pallets and hidden areas under equipment.

Methoprene is an insect growth regulator that mimics an insect's natural juvenile hormone. This chemical suppresses populations by impeding growth of individuals that it contacts, both by preventing maturity to the adult stage and causing reduced fecundity in those insects that are able to eclose. Methoprene has important applications in other stored product situations such grain bins (Chanbang et al. 2007) and other urban pest situations such as fire ant and mosquito control (Aubuchon et al. 2006, Henrick 2007). Stability over a wide range of

temperatures and has good residual activity are two of the characteristics that makes this chemical an attractive management option.

The usefulness of our economic analysis is that by standardizing costs to 283.7 m³ (10,000 ft³) gives way to rapidly calculate costs for facilities based on size. In our analysis, equipment costs were fixed, due to the unique needs and equipment requirements of different food manufacturing facilities and also shut-down time was fixed at two hours. Carrier costs can be especially important because methoprene is applied at a much lower rate (making amount of carrier higher); approximately seven times lower than the esfenvalerate, and is approximately eight times more expensive. Insect growth regulators can be expensive in comparison with conventional insecticides; it is important to be able to determine if increasing input costs by a small margin could give better control of stored pests or if it would be more cost effective to employ a different management option.

Especially with concerns about the safety of fumigants, the impending phase-out of methyl bromide and consumer preference for "safer" products, using insect growth regulators as control tactics is becoming more widespread. Management options such as methoprene for the control of Indianmeal moth provide good control while costing nearly the same as a more hazardous conventional insecticide. Studies such as this demonstrate that even difficult to kill insect pests may be effectively and economically managed with a lower risk insecticide. Though industrial usage of insect growth regulators may not be immediately adopted, alternatives to hazardous fumigants need to be made available.

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Figures and Tables

Figure 2-1 Experimental arenas used to contain wandering phase Indianmeal moth larvae. Construction of these units is described above. Drawings are not to scale.

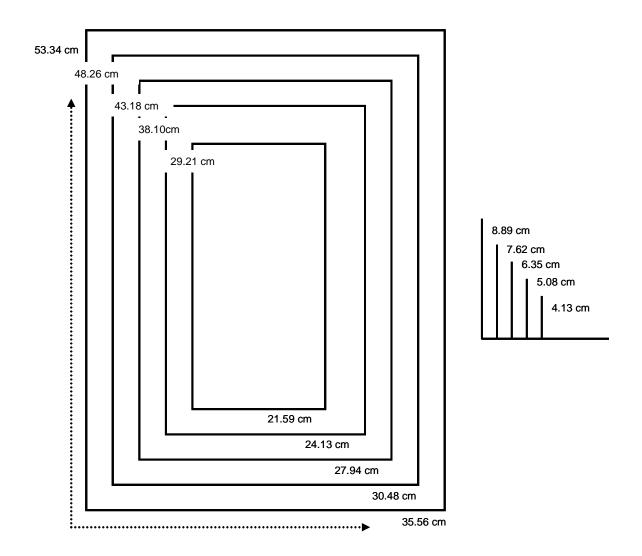


Figure 2-2 Experimental set-up of the pilot-scale sheds for all three simulated warehouse experiments. Concealed positions were created by making a scaled-down mock pallet described above. Position of the entry door is marked with the dashed gray curve and unobstructed and concealed positions are labeled. Drawings are not to scale.

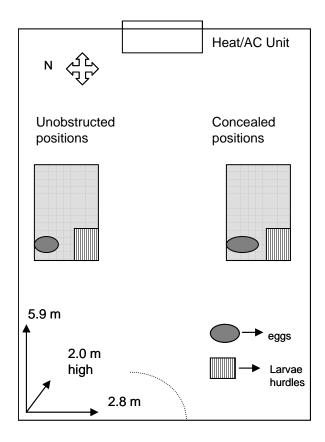


Figure 2-3 Survival of eggs and fifth instars of Indianmeal moth to the adult stage after exposure to aerosol methoprene treatments. Letters indicate statistical differences in means and standard errors using the MEANS procedure in SAS and means were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P=0.05 for each life stage. Average adult emergence was significantly different (P<0.0001) for each life stage between the methoprene treatments and the untreated control.

Untreated Control verses Label Rate Aerosol Methoprene Treatments

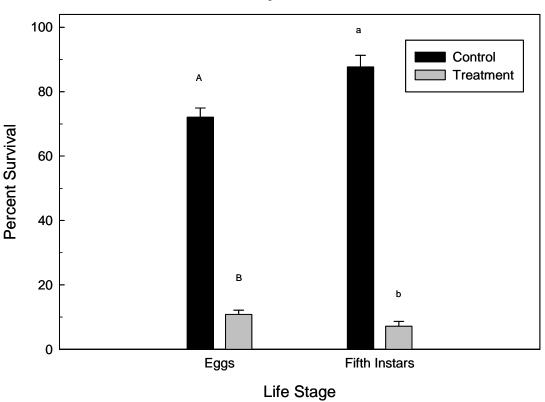
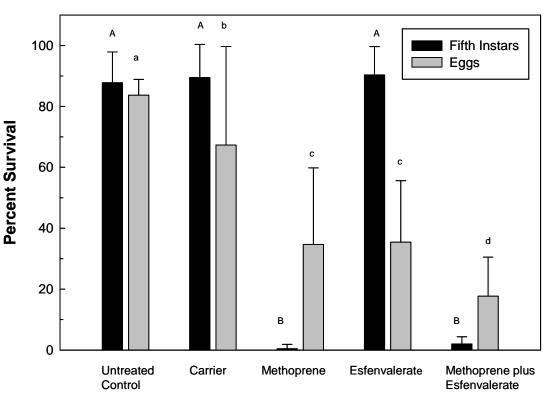


Figure 2-4 Comparison of survival of eggs and fifth instar Indianmeal moth to the adult stage exposed to aerosol methoprene and esfenvalerate treatments in pilot scale sheds. Capital letters indicate statistical differences between treatments for fifth instars and lower case letters indicated statistical differences between treatments for exposed eggs. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each life stage. There was no statistical difference in response to unobstructed or concealed positions (P = 0.1309).

Comparison of Aerosol Methoprene and Esfenvalerate Treatments by Life Stage



Treatments

Figure 2-5 Survival of eggs and fifth instar Indianmeal moth exposed to combinations of aerosol methoprene and esfenvalerate treatments at less than the label rate. Capital letters indicate statistical differences between treatments for fifth instars and lower case letters indicated statistical differences between treatments for exposed eggs. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each life stage. There was no statistical difference in response to unobstructed or concealed positions (P = 0.9170).

Comparison of Combination Treatments at Variable Rates

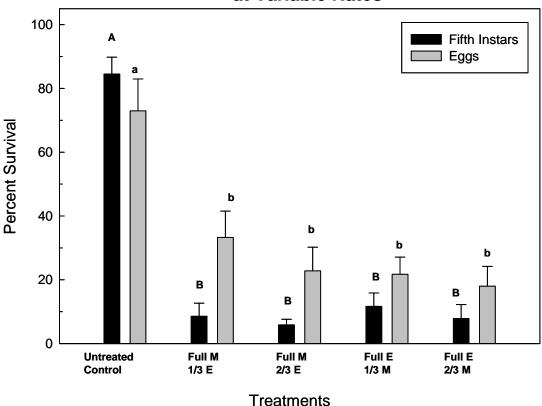


Table 2-1 Analysis of variance statistical effects on mortality of Indianmeal moth eggs and larvae exposed to aerosol methoprene treatments in unobstructed and concealed positions.

Effect	Num DF	Den DF	F Value	Pr > F
Treatment 1 (methoprene, control)	1	16.6	1037.08	<.0001
Treatment 2 (open, concealed)	1	16.6	1.29	0.2713
Treatment 1 * Treatment 2	1	16.6	0.00	0.9902
Life Stage (egg, larvae)	1	16.6	7.38	0.0149
Life Stage * Treatment 1	1	16.6	19.29	0.0004
Life Stage * Treatment 2	1	16.6	1.28	0.2736
Life Stage * Treatment 1 * Treatment 2	1	16.6	0.15	0.7009

Table 2-2 Analysis of variance statistical effects on mortality of Indianmeal moth eggs and larvae exposed to aerosol methoprene alone, aerosol esfenvalerate alone and both chemicals in combination treatments in unobstructed and concealed positions.

Effect	Num DF	Den DF	F Value	Pr > F
Treatment 1 (open, concealed)	1	95	0.01	0.9170
Treatment 2 (methoprene, control)	4	95	108.06	<.0001
Treatment 1 * Treatment 2	4	95	0.52	0.7230
Life Stage (egg vs. larvae)	1	95	4.30	0.0408
Life Stage * Treatment 1	1	95	1.67	0.1989
Life Stage * Treatment 2	4	95	26.12	<.0001
Life Stage * Treatment 1 * Treatment 2	4	95	0.09	0.9853

Table 2-3 Analysis of variance statistical effects on mortality of Indianmeal moth eggs and larvae exposed to aerosol methoprene and esfenvalerate combination treatments at four rates in unobstructed and concealed positions.

0.1309
<.0001
0.7027
0.0071
0.0381
0.0330
0.9159

Table 2-4 Summary of costs and risk levels for chemical treatments in Experiment 1 (methoprene vs. untreated control). Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticide and carrier oil costs as explained above. The scenario that represents the lowest risk at any threshold for both life stages is the methoprene insecticide treatment.

Economic Summary			
Costs:	Methoprene plus carrier	0.70	
per 10,000 ft ³			

Stage	Treatment	% Mortality	Risk (90%)	Risk (95%)	Risk (99%)	Cost (\$)
Eggs	Control	27.92 ^A	0.62	0.67	0.71	0.00
Eggs	Methoprene	89.14 ^B	0.04	0.07	0.10	0.70
Larvae	Control	12.26 ^a	0.78	0.83	0.87	0.00
Larvae	Methoprene	92.86 ^b	0.03	0.05	0.07	0.70

Table 2-5 Summary of costs and risk levels for chemical treatments in Experiment 2 (methoprene alone, esfenvalerate alone and combination treatment). Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticide and carrier oil costs as explained above. The scenario that represents the lowest risk at any threshold for both life stages is the methoprene and esfenvalerate combination treatment insecticide treatment.

Economic	c Summary													
Costs: per 10,000 ft ³		Carrier only Methoprene plus carrier Esfenvalerate alone Combination treatment		\$0.25 \$0.70 \$0.71 \$1.17										
								Stage	Treatment	% Mortality	Risk (90%)	Risk (95%)	Risk (99%)	Cost (\$)
								Eggs	Untreated Control	16.33 ^a	0.74	0.79	0.83	0.00
								Eggs	Carrier Only	32.71 ^b	0.59	0.63	0.66	0.25
Eggs	Methoprene Only	65.33°	0.26	0.30	0.34	0.70								
Eggs	Esfenvalerate Only	64.58°	0.26	0.31	0.34	0.71								
Eggs	Methoprene plus Esfenvalerate	82.33 ^d	0.10	0.13	0.17	1.17								
Larvae	Control	12.23 ^A	0.78	0.83	0.87	0.00								
Larvae	Carrier Only	9.34 ^A	0.79	0.84	0.88	0.25								
Larvae	Methoprene Only	99.58^{B}	0.00	0.00	0.00	0.70								

Larvae	Esfenvalerate Only	9.67 ^A	0.80	0.85	0.89	0.71
Larvae	Methoprene plus Esfenyalerate	98.01 ^B	0.00	0.00	0.01	1.17

Table 2-6 Summary of costs and risk levels for chemical treatments in Experiment 3. Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticide and carrier oil costs as explained above. The scenario that represents the lowest risk at any threshold for eggs is the full esfenvalerate + two-thirds methoprene treatment. For fifth instars the lowest risk treatment is full methoprene + two-thirds esfenvalerate.

Economic S	ummary					_
Costs: per 10,000 ft ³ .		Carrier only		\$0.25		_
		Methoprene plus carrier		\$0.70		
		Esfenvalerate alon	\$0.71			
		Combination treatment		\$1.17		
Stage	Treatment	% Mortality Ri		Risk	Risk	Cost (\$)
			(90%)	(95%)	(99%)	
Eggs	Untreated Control	27.07^{A}	0.63	0.68	0.72	0.00
Eggs	Full Esfenvalerate +	82.02^{B}	0.12	0.14	0.17	1.01
	2/3 Methoprene					
Eggs	Full Esfenvalerate +	78.30^{B}	0.14	0.18	0.21	0.85
	1/3 Methoprene					
Eggs	Full Methoprene +	77.20^{B}	0.15	0.19	0.22	1.00
	2/3 Esfenvalerate					
Eggs	Full Methoprene +	66.70^{B}	0.25	0.29	0.32	0.86
	1/3 Esfenvalerate					
Larvae	Untreated Control	15.48 ^a	0.75	0.80	0.84	0.00
Larvae	Full Esfenvalerate +	92.15 ^b	0.04	0.05	0.07	1.01
	2/3 Methoprene					
Larvae	Full Esfenvalerate +	88.33 ^b	0.06	0.08	0.11	0.85
	1/3 Methoprene					
Larvae	Full Methoprene +	94.15 ^b	0.01	0.03	0.05	1.00
	2/3 Esfenvalerate					
Larvae	Full Methoprene +	91.45 ^b	0.05	0.06	0.08	0.86
	1/3 Esfenvalerate					

CHAPTER 3 - Methoprene and Synergized Pyrethrins as an Aerosol Treatment of Commercial Buildings for the Management of Indianmeal moth (Lepidoptera: Pyralidae)

Abstract

New strategies for managing stored product pests are needed to fulfill the mandate to replace the fumigant methyl bromide in food manufacturing and storage facilities. Fumigant treatments in a commercial building can be costly in terms of insecticide costs and shut-down times. Aerosol insecticides delivered through an ultra-low volume application system provide one alternative, but little is known about the efficacy and distribution of these insecticides. One insecticide combination currently in use is synergized pyrethrins, alone and in combination with the insect growth regulator, methoprene. One stored product pest that is particularly difficult to manage is the Indianmeal moth (*Plodia interpunctella* Hübner). This insect causes contamination of stored products and sometimes equipment damage in food processing facilities. Infested products can also reach the consumer making management of the Indianmeal moth essential where food products are stored. There is little previous research on the efficacy of aerosol insecticides, especially methoprene, in the management of Indianmeal moth eggs. Eggs may be oviposited outside of food packages or in other exposed areas, making the egg stage a target for control as aerosols do not penetrate packaging materials as do fumigants. The study focused on two commercial buildings: a flour mill in Kansas and a grocery distribution warehouse in Missouri. The three goals were to: 1) compare the susceptibility of Indianmeal moth eggs when they are exposed to aerosol insecticide treatment, an Indianmeal moth diet that had been treated with insecticide, and packaging materials that had been exposed to aerosol insecticide; 2) evaluate the benefit of using a combination of methoprene and pyrethrin versus

methoprene only; and 3) examine the distribution of aerosol in "concealed" as well as "unobstructed" areas within a commercial facility.

Keywords: methoprene, esfenvalerate, packaging materials, Indianmeal moth

Introduction

The Indianmeal moth (*Plodia interpunctella* Hübner) can develop on a number of different commodities including grains, flours, beans, meals and dried fruits (Mohandass et al. 2007). For this reason it is a major pest in warehouses, elevators and commercial food processing facilities (Subramanyam and Hagstrum 1993, Doud and Phillips 2000, Arbogast et al. 2002). Indianmeal moths are specifically known to infest grain mills, as well as grocery facilities (Buchelos 1980, Campbell and Mullen 2004). Management of Indianmeal moth populations inside a food processing facility is essential because infestations of food materials during processing and of finished products lead to rejection of products by distribution centers and consumers (Campbell et al. 2002). Historically, many flour mills have relied on fumigation with methyl bromide to control stored product pests such as the Indianmeal moth; but this product is scheduled to be phased out under a world-wide agreement, the Montreal Protocol, (Anonymous 2004). Currently mills and process plants have received a critical use exemption (CUE) for the continued use of methyl bromide.

Because of the scheduled phase-out of methyl bromide, plus the fact that it is a very hazardous chemical, mills and processing facilities are evaluating safer and less expensive alternatives (Campbell et al. 2004, Mondall and Parween 2000). One available management option is to use an aerosol treatment with a mixture of the insect growth regulator, methoprene, and synergized pyrethrins. The advantage of this mixture is that the synergized pyrethrin product results in quick knock-down of the pests whereas the methoprene, which is stable over longer time periods, has residual activity (Mondall and Parween 2000). Both products are currently labeled for use in food handling facilities. Methoprene is registered by Central Sciences International (EPA reg. no. 2747-427); while several companies have registered their own proprietary formulations and application systems for synergized pyrethrins.

In determining what insect pest management methods to implement, cost and loss of production time are major considerations. Fumigants often require specialized training for application and can be expensive in terms of shut-down and applicator costs. An important aspect of any integrated pest management program is to determine the cost of a particular treatment method. Partial budget analysis can be used to compare chemical costs of control

strategies and can also be used to determine levels of risk associated with each strategy (Boehlje 1984). In the case of aerosol insecticide treatments, a partial budget analysis can compare different combinations of synergized pyrethrins and methoprene enabling food production plant managers and warehouse managers to be able to make better decisions. Economic analysis for some other methyl bromide alternatives using enterprise budgets for specific field crops have previously been developed (Nelson 1996, Byrd et al. 2006) and the same method can be applied to finished stored product situations.

Little research has been done on the efficacy of aerosol insecticide treatments in active commercial mills and warehouses, especially the use of insect growth regulators and their distribution as aerosols throughout a facility (Arthur 2008, Arthur and Campbell 2007). There is some previous laboratory research showing varying efficacy of synergized pyrethrins (both in direct contact and residual activity) for control of stored product beetles, but not for control of Indianmeal moth (Bernhard and Bennett 1981, Cline et al. 1984). Aerosol droplets may be impeded by barriers such as equipment, shelving and stored products. For this reason, information on distribution of the aerosol under such barriers must be obtained in order to determine the efficacy of aerosol treatments against the Indianmeal moth.

Indianmeal moth females oviposit eggs on a number of surfaces associated with food materials. Eggs may come into direct contact with aerosol particles, surfaces that have been exposed, and spilled food materials that have been exposed to aerosol particles. It is important to evaluate the efficacy of these chemicals in all three situations. The specific objectives of this experiment were to: 1) compare the susceptibility of Indianmeal moth eggs when they were exposed directly to aerosol insecticide treatment, various food materials that have been exposed to aerosol insecticides, and packaging materials that have been exposed to insecticides, 2) evaluate the benefit of using a combination of methoprene and pyrethrin versus methoprene-only and pyrethrin-only aerosol treatments, and 3) examine the distribution of aerosol particles in "concealed" and "unobstructed" areas as evidenced by survival of Indianmeal moth eggs to the adult stage.

Materials and Methods

General Procedures

A laboratory colony of Indianmeal moths was used for all studies. This colony was established in 1988 from individuals collected in Riley County, Kansas in the United States, and periodically supplemented with wild individuals. These colonies are maintained at the USDA-ARS Grain Marketing and Production Research Center in Manhattan, KS². The colony has been maintained in environmental growth chambers (Forma-Scientific, Thermo Electron Corporation, Waltham, MA) on an enriched wheat diet at 27±1°C, approximately 40% RH., and in darkness (L:D = 0:24 hrs). Relative humidity was maintained using pans of water in the bottom of the incubator. The colony was reared on a standard wheat-based diet. The standard rearing diet contains cracked wheat and shorts (4.4 kg), brewer's yeast (22g), sorbic and benzoic acid (9.5g each), honey (240ml), glycerin (240ml) and water (120ml).

For each study there were three treatment methods: 1) eggs exposed directly to settling aerosol particles, 2) diet materials exposed to the aerosol with unexposed eggs added following the treatment, and 3) packaging materials exposed to the aerosol with the corresponding diet inside, with unexposed eggs added following treatment. Eggs (less than 24 hours after oviposition) were exposed to the aerosol spray in 100 mm Petri dishes painted with black acrylic paint to reduce static. Eggs were transported to and from each facility in these dishes.

Approximately 100 g of each diet material were exposed to aerosols in 150 mm Petri dishes and after exposure diet materials were transferred to clean Petri dishes of the same size. Package materials with diets inside were placed directly on the floor of the facilities. After exposure, these materials were placed in clean plastic containers (disposable) purchased from the local grocery store that were 5 cm high, 13.7 cm wide, and 13.7 cm long. Following aerosol treatments, eggs that were exposed to insecticide were placed in clean diets and allowed to mature to the adult stage under the above conditions. Packaging and diet materials exposed to insecticide were transported back to the laboratory where eggs placed on these materials were allowed to mature to the adult stage under the previously described conditions. Lids on the 150

² Kansas State University Museum of Entomological and Prairie Arthropod Research under Lot Number 208

mm Petri dishes and the disposable plastic containers were altered to have a 5.08 cm (2 in) diameter vent cut in the top with a standard filter paper secured with a non-toxic adhesive to allow air movement. Temperature and relative humidity inside incubators were recorded using a HOBO data logger (Onset Computer Corporation, Bourne, MA).

Diet materials that were used for both studies were standard wheat-based diet (same as above), commercially available corn muffin mix and protein bars (peanut butter flavor) and raisins. Package material and diet combinations were standard wheat diet inside 16.8 cm x 14.9 cm sandwich-size zipper-seal plastic bags, commercially available peanut butter protein bar inside their original laminate paper packaging, corn muffin mix inside original packaging (laminated cardboard), and two snack-size boxes of raisins taped end to end inside original laminated cardboard packaging. All sizes of packaging materials were selected so that total surface area of exposure was approximately the same for all combinations. After exposure, we made small entry holes in each package (five holes in a star pattern) with a sharp probe to allow neonates to enter the packages.

Both field sites were equipped with an ultra-low-volume (ULV) compressed air application system, which dispensed aerosol insecticides at an approximate particle size of 15 microns, installed by Entech Systems (Kenner, Louisiana, USA). These application systems can be operated manually or by timer

Experimental Design for Flour Mill Experiment

The experiment was conducted in a commercial flour mill located in central Kansas. The mill was previously equipped a ULV system, through which a methoprene plus pyrethrin treatment is delivered on a regular basis. Additionally, there is an insect population sampling program and a temperature monitoring program ongoing in the facility. This mill actively practices integrated pest management (IPM) techniques such as sanitation as part of an overall pest management program. Materials for each block of the experiment were prepared and transported to the mill prior to each treatment period and transported back to the laboratory after exposure. One floor of the mill was chosen as the experimental site and controls were held in an unexposed building during fogging period. This floor of the mill had approximately 20-25% occupied space with milling equipment and was approximately 1,170 m² (18,480 ft²).

The experimental design was a split-split-split plot experiment blocked by time. The whole plot (WP) treatment was three replicated insecticide treatments: 1% pyrethrin plus methoprene, 3% pyrethrin only, and 3% pyrethrin plus methoprene. The 1% pyrethrin plus methoprene treatments were replicated four times with all sub-plot factors included and the 3% pyrethrin plus methoprene treatments replicated three times with exposed eggs only. The 3% pyrethrin only treatment was only run once and only because there was an error in shipping and the methoprene was not received at the mill in time to include it in the treatment. The 1% pyrethrin treatment was Entech Fog-10® (EPA Reg. No. 40391-10) which is 1.0 % pyrethrins, 2.0% piperonyl butoxide synergist, 3.33% N-octyl bicycloheptene dicarboximide and 93.67% inert ingredients. The 3% pyrethrin treatment was Entech Fog-30® (EPA Reg. No. 73049-400-40391) which contains 3.0 % pyrethrins, 6.0% piperonyl butoxide synergist, 10.0% N-octyl bicycloheptene dicarboximide and 81.0% inert ingredients. Each treatment was applied at the label rate for surface applications of 295.7 ml (10 oz) undiluted concentrate per 283.7 m³ (10,000 ft³). For the combination treatments, Diacon II[®] (methoprene) was added at the label rate for space applications, which is 900 mg of active ingredient [AI], per 283.7 m³ (10,000 ft³). For each test, treatments were placed in the mill and allowed to be exposed to the settling aerosol particles for two hours.

Sub-plot (SP) treatments were position in relation to spray nozzle (see Figure 1), "unobstructed" or "concealed" during treatment, method of treatment, and diet/packaging combinations. For this purpose of this study, "concealed" sites within each position were fully underneath a piece of equipment in the mill. While equipment in this mill was raised up off the floor, the treatments were still partially shielded from the aerosol particles as they settled, thus simulating food patches or insects under a pallet of product or equipment. "Unobstructed" sites were those not under equipment or other barriers. For each position and unobstructed/concealed sub-plot treatment, there were three exposure methods designed to simulate three possible scenarios. The purpose of this treatment was to compare the susceptibility of Indianmeal moth eggs (measured by emergence as adults) when they were exposed directly to aerosol insecticide treatment, exposed to food materials that had been treated with insecticide, and exposed to packaging materials that had been exposed to aerosol insecticide but which contained untreated food materials. The final sub-plot treatment was the type of diet/packaging material. Three representative package types were used: laminate paper, laminate cardboard and plastic. These

packages represent what would happen when damaged products were allowed to remain in the facility after insecticide treatment. Three types of diet were used for the diet exposure part of the study: standard Indianmeal moth diet, organic protein bar, and raisins. These products were chosen to represent spilled food material that could be found inside a commercial facility.

For each replicate, twenty eggs that had been exposed to the aerosol treatment were randomly chosen and placed in clean food material. In addition, twenty untreated eggs were placed on each of the exposed diets and on exposed packaging materials after they had been returned to the laboratory. Test dishes were held in a 27°C incubator with a pan of water as a humidity source (approximately 40% relative humidity). Adult emergence was recorded and experiments terminated when controls reached at least 75% adult emergence or when it was determined that there was no more survival which was approximately one week following beginning adult emergence from the control dishes. Beginning with block (time-replicate) 5, only eggs were exposed to the insecticide treatment, due to nearly zero adult emergence in the treated diet and treated packaging results from the previous time periods. Temperature and relative humidity inside incubators were recorded using a HOBO data logger (Onset Computer Corporation, Bourne, MA) placed next to one "unobstructed" and one "concealed" area during each treatment. A HOBO was also placed in the area where controls were being held. Temperatures and relative humidity in the mill fluctuated with the season. The experiment was run during a period from February 2007 through July 2007 and timing of application ranged from early morning to early evening. Temperatures ranged from 22±1 to 34±1°C and relative humidities from 23-65%.

Data were organized as a split-split plot design and analysis of variance was conducted using the PROC MIXED procedure in the Statistical Analysis System (SAS Institute 2001) with random effects were block (whole plot treatment) and block (whole plot treatment)*position. For percent survival to the adult stage for each life stage means and standard errors were also calculated using the MEANS Procedure of SAS. Means for treatments were separated using the Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.

Experimental Design for Grocery Warehouse Study

This experiment was conducted in a food distribution facility equipped with an aerosol fogging system (compressed air-operated) that is used infrequently to deliver methoprene and synergized pyrethrins. For these tests, Diacon II® was delivered through this system at the label rate (Central Sciences 2004) for space application which is 3 ml per 283.7 m³ (10,000 ft³). The test area (Figure 2) was a large, relatively open warehouse room with little fixed shelving. The room houses a variety of products (fresh and preserved food products, kitchen equipment and paper goods) that are moved in and out of the room frequently. The estimated occupied space (stacked goods) of this room at any given time is 10%. The volume of the room minus the 10% occupied space is approximately 15,050 m³. The room has multiple exits to the rest of the building, but no outside exits; therefore any insects moving into the room come from either the products or the adjacent warehouse. The treatment room has two nozzles for dispensing insecticide and the ULV system in this facility was installed such that one room can be treated separately from the others.

The experimental layout for this study was designed in a similar manner as the above experiment. Three sub-replicates in different areas of the fogged room were also done to reduce experimental variability, and controls were held in an office that was not exposed to the aerosol. The placement of packages was determined prior to the test. The same three package types were used as described in the previous experiment. For blocks (times) 1-4, standard wheat diet, protein bar and corn muffin mix were used; in blocks 5 and 6, raisins were substituted for the corn muffin mix due to low control survival. Packages were placed in the treated room and were considered either "open" or "concealed" in the same manner as in the previously-described experiment. This experiment was a split-split plot with the methoprene treatment as the whole plot factor blocked by time and the sub-plot as position in the warehouse. The sub-sub plot treatment was 2 areas ("unobstructed" or "concealed") within each position. Complications with the execution of this experiment created an imbalance in the experimental design: treated package data were available for the first two blocks only; corn muffin mix was used in blocks 3 and 4, and raisins were used in blocks 5 and 6. Treatments were placed in the room and the aerosol system started, then treatments were allowed to be exposed to the aerosol for two hours before collection. After the aerosol treatment, packages were removed from the room, placed in

plastic containers and transported back to our facility. There, eggs were added to the treatments and allowed to mature to the adult stage in the same manner described above.

Temperature and relative humidity inside incubators were recorded using a HOBO data logger (Onset Computer Corporation, Bourne, MA) placed next to treatment areas. All tests were conducted in the late afternoon during the summer of 2006; temperatures in the treatment area were $28 \pm 1^{\circ}$ C and relative humidities ranged from 30-70%. Temperatures in the control room were $24 \pm 1^{\circ}$ C with relative humidities of 50-60%. All data was analyzed for this experiment using the PROC MIXED procedure of the Statistical Analysis System (SAS Institute, 2001) in the same way as the previous experiment. Analysis was carried out on treated packaging materials in blocks 1 through 4 with results reported below. Due to the low control survival of treatments in blocks 3-4 in the other treatments, only blocks 5 and 6 are reported.

Partial Budget Analysis

Using chemical cost information calculated per 283.7 m³ (10,000 ft³), partial budget analysis revealed that to compare costs of methoprene and synergized pyrethrin treatments (alone and in combination) for two field sites for managing eggs of the Indianmeal moth. Risk was calculated at three levels (90, 95, and 99%) as deviations below the target goal. Tilley (2007) reports a modified Target MOTAD (mortality goal) model for optimizing cost and risk, but in the case of these three experiments time and equipment cost were fixed and the only variable cost was the cost of the chemical. Risk, in this case, was the inverse of mortality, up to the mortality target, at which it was set to zero, therefore the downside risk was mortality below the target level.

Chemical and carrier oil costs (for the methoprene only treatments) were calculated based on current industry prices. Carrier oil costs fluctuate with the global petroleum market, but for the purposes of this analysis they were fixed to \$0.83 per L (\$3.15 per gallon) or \$0.0008 per ml. Carrier costs can be especially important if comparisons are being made between chemicals with different application rates. In the case of the synergized pyrethrin applications, the formulation is premixed, so cost calculations were done on a per unit basis. For our analysis, means from all treatments in both the flour mill study and the grocery warehouse study were used to calculate overall mortality and risk. Since grocery warehouses are a mixture of products and package

types, we attempted to get an estimate of overall mortality and risk across all scenarios (treated eggs, treated packaging, and treated diet) for each diet type.

Results

Flour Mill Experiment

For blocks 1-4 (WP treatment 1% pyrethrin plus methoprene) analysis was run as if the control plates were an 11th position. Results are displayed in Table 3-1. Treating the control plates as a separate position was done because the controls were held in a separate room from the insecticide treatments. This procedure allowed us to better examine the effect of position within the treatment area. Unobstructed and concealed positions were not significantly different (P=0.7174). However, the method of treatment and diet had highly significant effect (P = <.0001). There were also significant differences among the three methods of treatment (treated eggs, treated package materials and treated diet) (Figures 3-5). When eggs were placed on any of the three treated diets, survival to the adult stage was virtually zero (Figure 3-3). Similarly, when eggs were exposed to treated packaging materials, adult emergence was also close to zero (Figure 3-5). However, while treatments were significantly different from controls (P=0.05), overall survival was lower when eggs were exposed directly to the aerosol treatment (Figure 3-4). Survival was significantly lower in the treatments involving raisins than in the other two diets. Because there was virtually no survival to adulthood of Indianmeal moths in both the treated diets and treated packages portion of the experiment, these factors were omitted from the final four blocks of the experiment.

Analysis of the treated egg treatment method across all blocks (1-8) and all three whole plot treatment methods (1% pyrethrin plus methoprene, 3% pyrethrin alone, and 3% pyrethrin plus methoprene) revealed no significant differences in the efficacy of treatments (P=0.5021) (Table 3-2). Analysis was conducted as described above with controls as position 11. The main effects diet and position were statistically significant at P<0.0001. Overall, treatments were significantly different from controls (P=0.05) but survival to the adult stage was higher than in the treated packaging and treated diet tests conducted in the first four blocks. Figure 3-6 displays differences by insecticide treatment and diet.

While assigning control tests to separate position made the overall position effect highly significant, it was important to determine if distance from the aerosol fogging nozzle in the treated tests was a significant factor as well. Figure 3-1 shows the positions in order of their approximate distance from the insecticide source and Table 3-3 shows the results of analysis of variance carried out on treated eggs for the effect of position on adult emergence. Treated diet and treated package data were omitted from this analysis because of the low survival to the adult stage in treatments. In general, as distance from the spray nozzle increased, so did survival to the adult stage (Figure 3-7).

Grocery Warehouse Study

Analysis was carried out in the same manner as the flour mill experiment. Table 3-4 shows the overall analysis of variance for treated package materials and diet combinations for blocks (times) 1-4. These were standard wheat diet inside plastic bags, commercially available peanut butter protein bar inside their original laminate paper packaging, and corn muffin mix inside original packaging (laminated cardboard). Analysis was done using control data as position 4 because the tests were held in an untreated room in the same facility (Table 3-4). Diet was the only main effect that was statistically significant (P<0.0001). Figure 8 shows the differences in diet/package type. The controls and treatments for wheat diet were significantly different at P=0.05, but not the corn muffin mix or the protein bar treatments and controls.

In blocks 3 and 4 there was low survival to the adult stage in controls of all three diets. Analysis of variance revealed no significant difference in positions or treatment type, though diet was still a significant main effect (P<0.0001) (Table 3-5). Possible reasons for the low survival include the handling of the treated eggs to and from the facility, escape from the plastic containers where the insects were allowed to mature, or nutritional quality of the wheat diet. Results are displayed in Figure 9. Analysis of variance is reported in Table 3-6 for blocks 5 and 6. Raisins were used instead of corn muffin mix in these two blocks due to higher adult emergence in the raisins. Table 3-7 shows means and standard errors for these two diets. Overall, the main effects of method of treatment and diet were statistically significant (P<0.0001). Results of means separation and differences by treatment method and diet are displayed in Figure 3-10.

Partial Budget Analysis

To make the best decisions in insect pest management, the cost of any given control method must be weighed against its efficacy. In the case of our partial budget analysis, efficacy was measured by overall mortality (failure to emerge as adults) Results of these are displayed in Tables 3-8 to 3-10. Methoprene was applied at approximately 100 times the rate of the synergized pyrethrin formulation (29.57 versus 295.7 ml per 10,000m³) and costs 8.5 times as much as the most expensive pyrethrin treatment tested (3%). Our results indicate that while the 3% pyrethrin alone reduced survival of eggs to the adult stage by 80.18%, methoprene alone only reduced adult emergence in eggs exposed directly to the aerosol treatment by 72.06%. The 1% combination treatment was approximately as effective as the 3% combination treatment; 83.06% and 78.39% respectively when comparing the treated egg method. See Table 3-10 for full comparison. When treated packaging and treated diet data are factored in, reduction in survival for the methoprene alone and 1% pyrethrin plus methoprene treatments was substantially increased but we can only compare the treated egg scenario across all insecticide treatments. Of all treatment scenarios, the 1% rate synergized pyrethrin plus methoprene represents the lowest risk, but not the highest cost. While the cost is still almost four times the cost of methoprene alone (\$3.14 versus \$0.71), mortality was significantly increased with the addition of the 1% synergized pyrethrin.

Discussion

These studies show that insecticides delivered via aerosols can be a feasible alternative to traditional methods such as fumigation, especially when considering chemical costs. Also, the installation of a facility-wide system may be less expensive than the cost of one fumigation treatment depending on size and specific needs. Previous research indicates that Indianmeal moth survival to the adult stage is prevented when larvae and eggs are fed diets exposed to methoprene (Arthur, unpublished data, Oberlander, et al., 1997). We showed the same results on four different diets. Indianmeal moth survival is also reduced when eggs and larvae are exposed to packaging materials or aerosol treatments of methoprene (Jenson et al., unpublished data). Although we would have expected to see better efficacy with the treatments in the open position because aerosol droplets may be impeded by equipment and other barriers, survival in unobstructed and concealed positions was not significantly different. Therefore this study also

demonstrates that aerosol particles will carry beneath stacked products and machinery in commercial environments. Our results also showed better efficacy and distribution of aerosol synergized pyrethrins than in previous experiments (see Bernhard and Bennett 1981, Cline et al. 1984). Our results for synergized pyrethrins are also similar to other laboratory studies involving grain (corn and wheat) treated with synergized pyrethrins that showed good efficacy against the rice moth (*Corcyra cephalonica*) and Indianmeal moth (Huang and Subramanyam 2003, 2005).

The efficacy of aerosol treatments with methoprene alone was similar to the pyrethrin plus methoprene treatments with respect to eggs exposed to treated packaging materials and to treated diets. Survival to the adult stage in these treatments overall was very low. However, eggs that were directly exposed to the methoprene-only aerosol treatment had a much higher survival to the adult stage than the pyrethrin alone treatments or the combination of the two insecticides (Figures 3-6 and 3-10). The overall feasibility of all aerosol treatments was demonstrated by the cost-risk analysis. Total mortality of the egg stage of the Indianmeal moth across all treatment scenarios was between 70 and 85%, so these treatments would be expected to significantly alter population dynamics of insects infesting these products. Additionally, our findings indicated that the most expensive treatment option is not always the lowest risk. For example, the 3% pyrethrin plus methoprene treatment did not reduce survival to the adult stage as much as the other treatments. These results represent the efficacy of these chemicals for eggs of the Indianmeal moth only. Consideration of Indianmeal moth life stages, other pest species and particular needs of each facility must be carefully assessed before insecticide application.

Field trials are especially important to conduct when attempting to assess insecticides that are currently being used in food-processing facilities. This study also adds to the growing information that reduced-risk insecticides in the form of aerosols can be combined with sampling and sanitation as part of a comprehensive pest management program. By utilizing other integrated pest management techniques such as sanitation, the risk of infestations in spilled food, such as in our treated diet scenario would decrease. Also, using pheromone trapping to monitor Indianmeal moth populations is a valuable tool in determining whether or not to treat the facility with insecticide (Doud and Phillips 2000, Campbell et al. 2002).

We had a unique opportunity to work closely with cooperators at these two commercial facilities to evaluate this integrated pest management alternative under "real-world" conditions. By working with mill managers and exchanging information, we were able to test previous

laboratory research on the chemical control of Indianmeal moth with methoprene in a large-scale environment. One limitation of field research of this nature is that normally live insects cannot be brought into a facility. We were able to assess eggs of the Indianmeal moth, but other life-stages should be assessed. Further research into methods of safely containing other life stages and insect species should be conducted so that overall population reduction can be assessed in large-scale experiments. Another limitation of using commercial field sites is that the timing of sprays, and concentrations and combinations used, are fully determined by the cooperator. Therefore, the ideal number of replications may not be reached, and other combinations of insecticides that are of interest may not necessarily be tested. Large-scale field studies in facilities, especially facilities that store finished stored products, are vital in understanding how efficacy differs from laboratory research and how pests can be managed safely in situations where food is stored for human consumption. Due to the complex nature of grocery warehouses (variation of product, frequent rotation of product, little control over product in transit) field experiments also help to clarify specific needs of industry in developing integrated pest management strategies.

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Figures and Tables

Figure 3-1 Flour Mill Experiment. Equipment and treatment layout of the flour mill experiment. The black oval represents the position of the aerosol delivery nozzle and dark gray blocks represent exits from that floor of the mill. Drawings are not to scale.

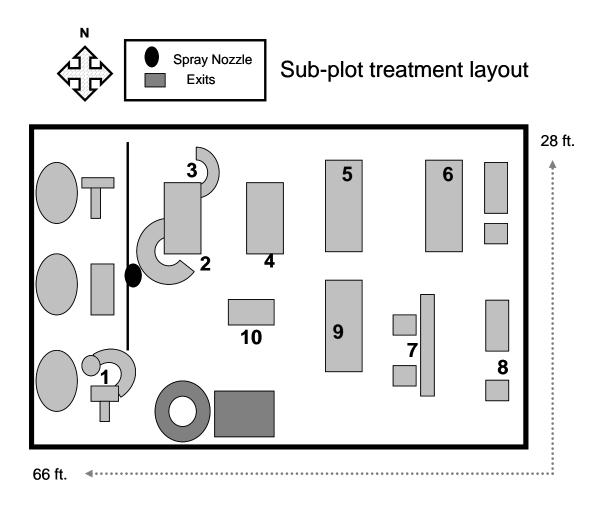


Figure 3-2 Grocery Distribution Warehouse Experiment. Dark gray circles represent locations of aerosol delivery nozzles. Breaks in the perimeter indicate exits from test room. Positions inside this room were chosen at random prior to testing periods. Locations of stacked product and shelving varied with each time period. Drawings are not to scale.

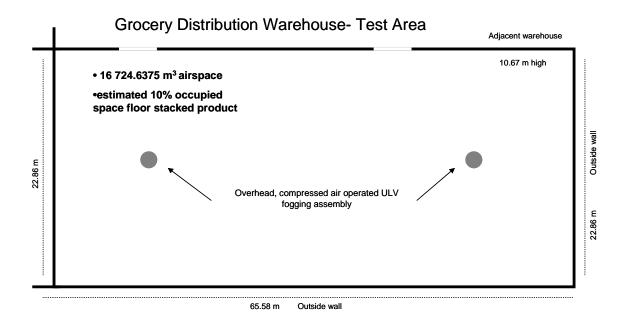


Figure 3-3 Flour Mill Experiment. Survival of Indianmeal moth eggs exposed to three diet types exposed to a 1% synergized pyrethrin plus methoprene aerosol treatment at the label rate for space applications. Capital letters indicate statistical differences between diet types and asterisks indicate statistical differences between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P =0.05 for each treatment combination.

Eggs Exposed to Treated Diets

Protein Bar Wheat Diet Raisins

*

0
20
40
60
80

Percent Survival to the Adult Stage

Figure 3-4 Flour Mill Experiment. Survival of Indianmeal moth eggs exposed directly to a 1% synergized pyrethrin plus methoprene aerosol treatment at the label rate for space applications and reared on three diet types. Capital letters indicate statistical differences between diet types and lower case letters indicate statistical differences between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P=0.05 for each treatment combination.

Eggs Directly Exposed to the Aerosol Treatment

Protein Bar Wheat Diet Raisins

A

O 20 40 60 80

Percent Survival to the Adult Stage

82

Figure 3-5 Flour Mill Experiment. Survival of Indianmeal moth eggs exposed to three diet/package type combinations exposed to a 1% synergized pyrethrin plus methoprene aerosol treatment at the label rate for space applications. Capital letters indicate statistical differences between diet types and asterisks indicate statistical differences between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.

Eggs Exposed to Treated Packaging Materials

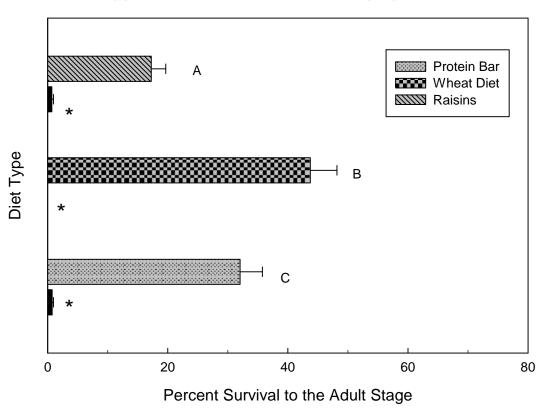


Figure 3-6 Flour Mill Experiment. Survival of Indianmeal moth eggs exposed directly to three aerosol insecticide combinations (1% synergized pyrethrin plus methoprene, 3% synergized pyrethrin alone, and 3% synergized pyrethrin plus methoprene) Asterisks indicate statistical differences among treatments and between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.

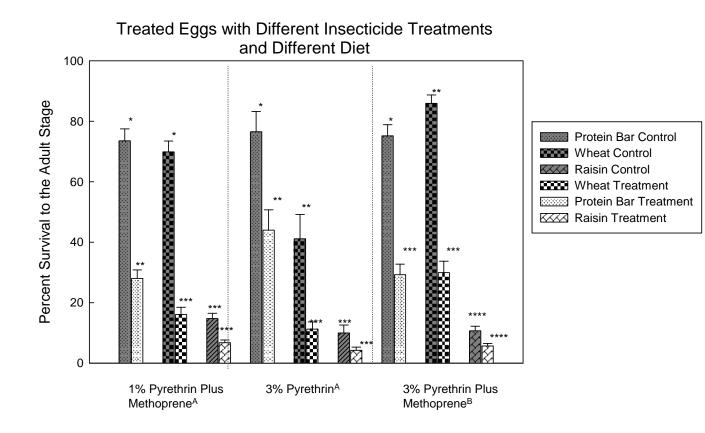


Figure 3-7 Flour Mill Experiment. Survival to the adult stage for Indianmeal moth eggs exposed to three aerosol insecticide combinations (1% synergized pyrethrin plus methoprene, 3% synergized pyrethrin alone, and 3% synergized pyrethrin plus methoprene). Treatments were pooled and overall means plotted. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination. Lower case letters indicate differences in adult emergence by position.

Positions at Varying Distance from Spray Nozzle

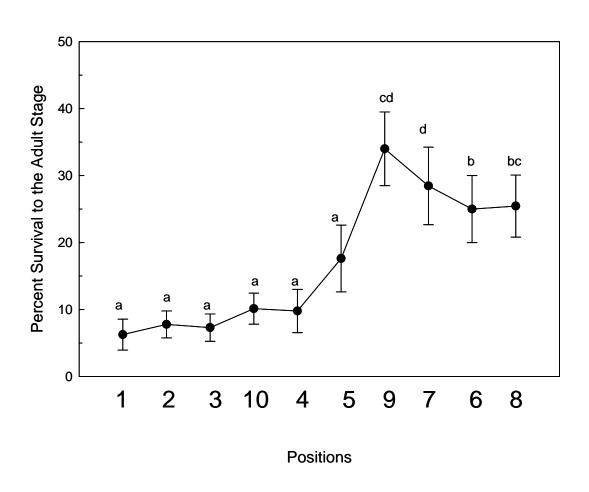
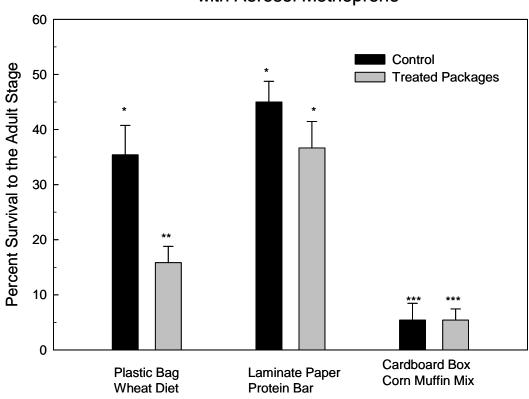


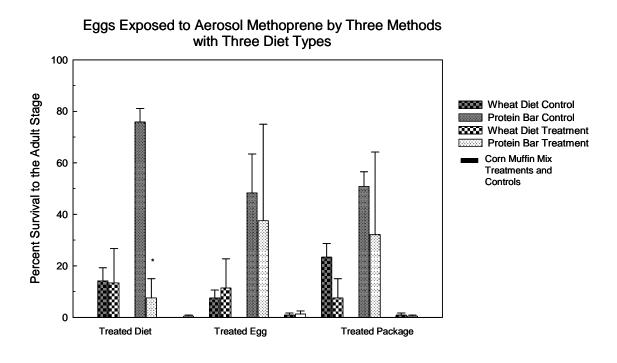
Figure 3-8 Grocery Distribution Warehouse Experiment. Survival of Indianmeal moth eggs exposed to three diet/package type combinations exposed to an aerosol methoprene treatment at the label rate for space applications. Asterisks indicate statistical differences among treatments and between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P=0.05 for each treatment combination.

Eggs Exposed to Three Package and Diet Types Treated with Aerosol Methoprene



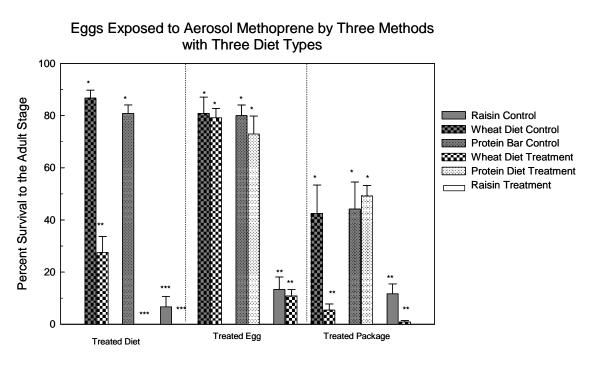
Package and Diet Type

Figure 3-9 Grocery Distribution Warehouse Experiment. Blocks 3 and 4. Survival of Indianmeal moth eggs exposed to three treatment methods (treated diet, treated eggs, treated packaging exposed to a methoprene aerosol treatment at the label rate for space applications. Asterisks indicate statistical differences among treatments and between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.



Treatment Method

Figure 3-10 Grocery Distribution Warehouse Experiment. Blocks 5 and 6. Survival of Indianmeal moth eggs exposed to three treatment methods (treated diet, treated eggs, treated packaging exposed to a methoprene aerosol treatment at the label rate for space applications. Asterisks indicate statistical differences among treatments and between treatments and controls. Mean and standard errors were calculated using the MEANS procedure in SAS and means were separated using Ryan-Einot-Gabriel-Welsch Multiple Range test at a significance level of P = 0.05 for each treatment combination.



Treatment Method

Table 3-1 Flour Mill Experiment. Results of analysis of variance of survival to the adult stage of Indianmeal moth eggs exposed to three treatment methods (treated diet, treated eggs, treated packaging) during a 1% synergized pyrethrin plus methoprene aerosol treatment at the label rate for space applications. Treatments are labeled below. Main effects that were significant at the P=0.05 level were treatment method and diet. There were also several significant main effect interactions.

Effect	Num DF	Den DF	F Value	Pr > F
Whole Plot Treatment 1% Pyrethrin and Methoprene				
Position (1-10 treated, 11 control)	9	30	1.01	0.4528
(sub-plot)				
Treatment 1(egg, pack, diet)	2	849	116.73	<.0001
(sub-sub-sub plot)				
Position * Treatment 1	18	849	3.19	<.0001
Treatment 2 (unobstructed, concealed) (sub-sub plot)	1	849	0.13	0.7174
Position * Treatment 2	9	849	0.28	0.9792
Treatment 1 * Treatment 2	2	849	0.59	0.5557
Position * Treatment 1 * Treatment 2	18	849	0.27	0.9990
Diet	2	849	63.42	<.0001
Position * Diet	18	849	0.58	0.9148
Treatment 1 * Diet	4	849	18.74	<.0001
Position * Treatment 1 * Diet	36	849	0.71	0.9030
Treatment 2 * Diet	2	849	0.29	0.7472
Position * Treatment 2 * Diet	18	849	0.27	0.9989
Treatment 1 * Treatment 2 * Diet	4	849	0.14	0.9677
Position * Treatment 1 * Treatment 2 * Diet	36	849	0.29	1.0000

Table 3-2 Flour Mill Experiment. Results of analysis of variance of survival to the adult stage of Indianmeal moth eggs exposed to three aerosol insecticide combinations (1% synergized pyrethrin plus methoprene, 3% synergized pyrethrin alone, and 3% synergized pyrethrin plus methoprene (at the label rate for space applications)) and reared on three diet types. Main effects that were significant at the P=0.05 level were position and diet. There were also several significant main effect interactions.

Effect	Num	Den DF	F Value	Pr > F
	DF			
Whole Plot Treatment	2	5	0.79	0.5021
(1%, 3% Pyrethrin plus Methoprene, Methoprene only)				
Position (1-10 treated, 11 control)	9	50	12.29	<.0001
(sub-plot)				
Whole Plot Treatment * Position	18	50	1.38	0.1822
Treatment 2 (unobstructed, concealed)	1	467	0.07	0.7967
(sub-sub- plot)				
Whole Plot Treatment * Treatment 2	2	467	0.99	0.3730
Position * Treatment 2	9	467	0.40	0.9333
Whole Plot Treatment * Position * Treatment 2	18	467	0.56	0.9262
Diet	2	467	174.67	<.0001
Whole Plot Treatment * Diet	4	467	15.92	<.0001
Position * Diet	18	467	4.38	<.0001
Whole Plot Treatment * Position * Diet	36	467	1.22	0.1870
Treatment 2* Diet	2	467	0.34	0.7138
Whole Plot Treatment * Treatment 2* Diet	4	467	0.15	0.9608
Position * Treatment 2 * Diet	18	467	0.43	0.9805
Whole Plot Treatment * Position * Treatment 2 * Diet	35	467	0.68	0.9170
Diet	2	467	174.67	<.0001
Whole Plot Treatment * Diet	4	467	15.92	<.0001
Position * Diet	18	467	4.38	<.0001
Whole Plot Treatment * Position * Diet	36	467	1.22	0.1870
Treatment 2 * Diet	2	467	0.34	0.7138
Whole Plot Treatment * Treatment 2 * Diet	4	467	0.15	0.9608
Position * Treatment 2 * Diet	18	467	0.43	0.9805
Whole Plot Treatment * Position * Treatment 2* Diet	35	467	0.68	0.9170

Table 3-3 Flour Mill Experiment. Results of analysis of variance of mortality of Indianmeal moth eggs and larvae exposed to aerosol treatments in unobstructed and concealed positions. Indianmeal moth eggs were exposed to three aerosol insecticide combinations (1% synergized pyrethrin plus methoprene, 3% synergized pyrethrin alone, and 3% synergized pyrethrin plus methoprene) at the label rate for space application. Treatments were pooled and overall means plotted.

Effect	Num	Den DF	F Value	Pr > F
Position Effect, Treated Eggs	DF			
Whole Plot Treatment	2	5	0.74	0.5210
(1%, 3% Pyrethrin plus Methoprene , Methoprene only)				
Position (1-10 treated, 11 control)	10	50	23.54	<.0001
(sub-plot)				
Whole Plot Treatment * Position	20	50	1.43	0.1518
Diet	2	556	134.31	<.0001
Whole Plot Treatment * Diet	4	556	13.92	<.0001
Position * Diet	20	556	11.24	<.0001
Whole Plot Treatment * Position * Diet	40	556	1.34	0.0856

Table 3-4 Grocery Distribution Warehouse Experiment. Results of analysis of variance of mortality of Indianmeal moth eggs exposed to package materials and diet combinations that were exposed to aerosol methoprene treatments in unobstructed and concealed positions. Main effects that were significant at the P=0.05 level were diet. There were no significant main effect interactions.

Effect	Num DF	Den DF	F Value	Pr > F
Position	2	9	1.29	0.3223
(1-3 treated, 4 control) (sub-plot)	2	9	1.29	0.3223
Diet	2	75	40.95	<.0001
Position * Diet	4	75	0.66	0.6203
Treatment 2 (unobstructed, concealed)	1	75	0.01	0.9392
(sub-sub- plot)	1	73	0.01	0.9392
Position * Treatment 2	2	75	0.29	0.7482
Treatment 2 * Diet	2	75	0.43	0.6539
Position * Treatment 2 * Diet	4	75	0.39	0.8152

Table 3-5 Grocery Distribution Warehouse Experiment Results of analysis of variance of mortality of Indianmeal moth eggs exposed to three methods (treated diet, treated eggs, treated packaging) that were exposed to aerosol methoprene treatments in unobstructed and concealed positions. Main effects that were significant at the P=0.05 level were diet. There were no significant main effect interactions all involving diet.

Effect	Num DF	Den DF	F Value	Pr > F
Position	2	3	0.63	0.5899
(1-3 treated, 4 control) (sub-plot)	2	3	0.03	0.3077
Treatment 1	1	94	2.46	0.1198
(unobstructed, concealed) (sub-sub- plot)	1	71	2.10	0.1170
Position * Treatment 1	2	94	0.97	0.3828
Treatment 2	2	94	2.00	0.1410
(egg, pack, diet) (sub-sub-sub plot)	2	94	2.00	0.1410
Position * Treatment 2	4	94	2.53	0.0458
Treatment 1 * Treatment 2	2	94	0.67	0.5162
Position * Treatment 1 * Treatment 2	4	94	1.70	0.1575
Diet	2	94	10.29	<.0001
Position * Diet	4	94	1.83	0.1297
Treatment 1 * Diet	2	94	1.21	0.3019
Position * Treatment 1 * Diet	4	94	1.52	0.2022
Treatment 2 * Diet	4	94	0.91	0.4628
Position * Treatment 2 * Diet	8	94	1.53	0.1588
Treatment 1 * Treatment 2 * Diet	4	94	1.54	0.1972
Position*Treatment 1*Treatment 2*Diet	8	94	1.47	0.1770

Table 3-6 Grocery Distribution Warehouse Experiment Results of analysis of variance of mortality of Indianmeal moth eggs exposed to three methods (treated diet, treated eggs, treated packaging) that were exposed to aerosol methoprene treatments in unobstructed and concealed positions. Main effects that were significant at the P=0.05 level were diet. There were several significant main effect interactions all involving diet.

Effect	Num DF	Den DF	F Value	Pr > F
Position	2	3	1.40	0.3720
(1-3 treated, 4 control) (sub-plot)	2	3	1.40	0.3720
Treatment 1	1	95	0.93	0 2201
(unobstructed, concealed) (sub-sub- plot)	1	93	0.93	0.3381
Position * Treatment 1	2	95	1.38	0.2557
Treatment 2	2	0.5	116.22	< 0001
(egg, pack, diet) (sub-sub-sub plot)	2	95	116.23	<.0001
Position * Treatment 2	4	95	2.28	0.0665
Treatment 1 * Treatment 2	2	95	0.59	0.5543
Position * Treatment 1 * Treatment 2	4	95	0.40	0.8065
Diet	2	95	174.19	<.0001
Position * Diet	4	95	3.26	0.0151
Treatment 1 * Diet	2	95	0.12	0.8828
Position * Treatment 1 * Diet	4	95	1.84	0.1275
Treatment 2 * Diet	4	95	36.47	<.0001
Position * Treatment 2 * Diet	8	95	0.98	0.4539
Treatment 1 * Treatment 2 * Diet	4	95	0.27	0.8987
Position*Treatment 1*Treatment 2*Diet	8	95	1.42	0.1992

Table 3-7 Grocery Distribution Warehouse Experiment. Means and standard error generated using SAS for the substitution of raisins for corn muffin mix in the final two blocks of the experiment.

Treatment Method	Methoprene Treatment	Diet Type	Mean	Std. Error (±)
Treated Diet	Control	Corn muffin mix	0	0
Treated Eggs	Control	Corn muffin mix	0.83	0.83
Treated Diet	Label Rate	Corn muffin mix	0.417	0.417
Treated Eggs	Label Rate	Corn muffin mix	1.25	0.65
Treated Diet	Control	Raisins	6.67	4.01
Treated Eggs	Control	Raisins	13.33	4.77
Treated Diet	Label Rate	Raisins	0	0
Treated Eggs	Label Rate	Raisins	10.83	2.45

Table 3-8 Summary of costs and risk levels for chemical treatments in Flour Mill Experiment (1% synergized pyrethrin plus methoprene, 3% synergized pyrethrin alone, and 3% synergized pyrethrin plus methoprene) versus untreated controls. Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticides as explained above. Treated eggs are compared across all treatment levels. The scenario that represents the lowest risk, lowest cost and highest mortality is the 1% pyrethrin plus methoprene treatment.

1% Pyrethrin Plus	Treatment Method Chemical Treatmen	t % Mortality	Risk	Risk	Risk	Cost (\$)
	Aerosol treatment- 3% Pyrethrin	80.18%	12.99%	15.82%	19.02%	\$5.39
Total	No treatment	57.46%	33.91%	38.06%	41.64%	\$0.00
Raisins	3% Pyrethrin Only	95.75%	0.0050	0.0125	0.0365	\$5.39
Wheat diet	3% Pyrethrin Only	88.75%	0.0425	0.0725	0.1045	\$5.39
Protein bar	3% Pyrethrin Only	56.05%	0.3421	0.3895	0.4295	\$5.39
Raisins	Control/None	90.00%	0.0300	0.0600	0.0920	\$0.00
Wheat diet	Control/None	58.89%	0.3222	0.3667	0.4022	\$0.00
Protein bar	Control/None	23.50%	0.6650	0.7150	0.7550	\$0.00
Diet	Chemical Treatment	70 19101 tuttiy	(90%)	(95%)	(99%)	υσι (φ)
3% Pyrethrin Only	Treatment Method- Treated Eggs	% Mortality	Risk	Risk	Risk	Cost (\$)
Summaries by Chemi	cal Combination					
	3% pyrethrin plus methoprene	\$ 5.851				
	1% pyrethrin plus methoprene	\$ 3.141				
Costs: per 10,000 ft ³	3% pyrethrin only	\$ 5.390				
Economic Summary						

Methoprene				(90%)	(95%)	(99%)	
Diet				(90%)	(9370)	(9970)	
Protein bar	Treated Diet	Control/None	45.38%	0.4750	0.5088	0.5388	\$0.00
Wheat diet	Treated Diet	Control/None	35.88%	0.5613	0.6000	0.6330	\$0.00
Raisins	Treated Diet	Control/None	90.50%	0.0400	0.0650	0.0890	\$0.00
Protein bar	Treated Diet	1% Pyrethrin plus Methop.	98.50%	0.0063	0.0088	0.0138	\$3.14
Wheat diet	Treated Diet	1% Pyrethrin plus Methop.	99.81%	0.0000	0.0006	0.0016	\$3.14
Raisins	Treated Diet	1% Pyrethrin plus Methop.	100.00%	0.0000	0.0000	0.0000	\$3.14
Protein bar	Treated Eggs	Control/None	26.47%	0.6403	0.6878	0.7258	\$0.00
Wheat diet	Treated Eggs	Control/None	30.17%	0.5983	0.6483	0.6883	\$0.00
Raisins	Treated Eggs	Control/None	85.28%	0.0760	0.1110	0.1400	\$0.00
Protein bar	Treated Eggs	1% Pyrethrin plus Methop.	72.00%	0.2005	0.2370	0.2714	\$3.14
Wheat diet	Treated Eggs	1% Pyrethrin plus Methop.	83.94%	0.1059	0.1298	0.1544	\$3.14
Raisins	Treated Eggs	1% Pyrethrin plus Methop.	93.24%	0.0183	0.0375	0.0616	\$3.14
Protein bar	Treated Packages	Control/None	67.95%	0.2462	0.2821	0.3128	\$0.00
Wheat diet	Treated Packages	Control/None	56.25%	0.3538	0.3950	0.4290	\$0.00
Raisins	Treated Packages	Control/None	82.75%	0.1050	0.1375	0.1655	\$0.00
Protein bar	Treated Packages	1% Pyrethrin plus Methop.	99.24%	0.0000	0.0000	0.0060	\$3.14
Wheat diet	Treated Packages	1% Pyrethrin plus Methop.	100.00%	0.0000	0.0000	0.0000	\$3.14
Raisins	Treated Packages	1% Pyrethrin plus Methop.	99.30%	0.0006	0.0019	0.0059	\$3.14
Total	No treatment*		47.31%	43.82%	48.23%	51.80%	\$0.00
	Aerosol 1% Pyreth	nrin plus Methoprene*	83.06%	10.82%	13.48%	16.25%	\$3.14
	* Treated Eggs only	y for comparison with other					
	chemical treatments	S					
3% Pyrethrin plus			a. 3.5	Risk	Risk	Risk	C = = 4 (th)
Methoprene	Treatment Method- Treated Eggs		% Mortality	(90%)	0%) (95%)	(99%)	<i>Cost</i> (\$)

Diet	Chemical Treatment					
Protein bar	Control/None	24.83%	0.6550	0.7033	0.7420	\$0.00
Wheat diet	Control/None	14.14%	0.7586	0.8086	0.8486	\$0.00
Raisins	Control/None	89.31%	0.0328	0.0603	0.0976	\$0.00
Protein bar	3% Pyrethrin plus Methoprene	70.77%	0.2123	0.2498	0.2838	\$5.85
Wheat diet	3% Pyrethrin plus Methoprene	70.08%	0.2233	0.2567	0.2907	\$5.85
Raisins	3% Pyrethrin plus Methoprene	94.32%	0.0119	0.0254	0.0505	\$5.85
Total	No treatment	42.76%	48.21%	52.41%	56.27%	\$0.00
	Aerosol 3% Pyrethrin plus Methoprene	78.39%	14.92%	17.73%	20.83%	\$5.85

Table 3-9 Grocery Distribution Warehouse Experiment. Summary of costs and risk levels for chemical treatments by treatment method for aerosol methoprene alone versus untreated controls. Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticide and carrier cost as explained above. Treated eggs, treated packaging and treated diet are compared across all diets. The scenario that represents the lowest risk, lowest cost and highest mortality is the treated diet method for all diets.

Economic Summary							
Costs: per 10,000 ft ³	Methoprene only	\$ 0.461					
	Plus carrier cost	\$ 0.711					
Summaries by Treatm	nent Method						
			0/ Montality	Risk	Risk	Risk	Cost (\$)
Treated Diet	Chemical Treatment		% Mortality	(90%)	(95%)	(99%)	<i>Cost</i> (\$)
Corn Mix	Control/None		100.00%	0.0000	0.0000	0.0000	\$0.00
Protein Bar	Control/None		21.67%	0.6833	0.7333	0.7733	\$0.00
Raisins	Control/None		93.33%	0.0250	0.0417	0.0617	\$0.00
Wheat diet	Control/None		49.58%	0.4208	0.4625	0.4958	\$0.00
Corn Mix	Label Methoprene		99.58%	0.0000	0.0000	0.0033	\$0.71
Protein Bar	Label Methoprene		96.25%	0.0208	0.0250	0.0350	\$0.71
Raisins	Label Methoprene		100.00%	0.0000	0.0000	0.0000	\$0.71
Wheat diet	Label Methoprene		0.80	0.12	0.16	0.20	\$0.71
Total	No treatment		66.15%	28.23%	30.94%	33.27%	\$0.00
	Aerosol methoprene		93.85%	3.49%	4.58%	5.83%	\$0.71
			0/ Mantalita	Risk	Risk	Risk	C = -4 (\$)
Treated Eggs	Chemical Treatment		% Mortality	(90%)	(95%)	(99%)	<i>Cost</i> (\$)

Corn Mix	Control/None	99.17%	0.0000	0.0000	0.0067	\$0.00
Protein Bar	Control/None	35.83%	0.5542	0.5958	0.6325	\$0.00
Raisins	Control/None	88.00%	0.0600	0.0900	0.1140	\$0.00
Wheat diet	Control/None	57.69%	0.3423	0.3808	0.4146	\$0.00
Corn Mix	Label Methoprene	99.09%	0.0000	0.0000	0.0073	\$0.71
Protein Bar	Label Methoprene	44.79%	0.4667	0.5083	0.5433	\$0.71
Raisins	Label Methoprene	89.17%	0.0333	0.0708	0.1008	\$0.71
Wheat diet	Label Methoprene	52.21%	0.3729	0.4083	0.44	\$0.71
Total	No treatment	70.17%	23.91%	26.67%	29.19%	\$0.00
	Aerosol methoprene	72.06%	21.82%	24.69%	27.29%	\$0.71
		- 0/35 . 10	Risk	Risk	Risk	~ .d\
Treated Packages	Chemical Treatment	% Mortality	(90%)	(95%)	(99%)	<i>Cost</i> (\$)
laminate cardboard	Control/None	93.75%	0.0208	0.0375	0.0575	\$0.00
laminate paper	Control/None	52.50%	0.3750	0.4250	0.4650	\$0.00
plastic bag	Control/None	67.08%	0.2292	0.2792	0.3192	\$0.00
laminate cardboard	Label Methoprene	99.38%	0.0000	0.0000	0.0050	\$0.71
laminate paper	Label Methoprene	59.38%	0.3146	0.3604	0.3971	\$0.71
plastic bag	Label Methoprene	93.54%	0.0208	0.0396	0.0596	\$0.71
Total	No treatment	71.11%	20.83%	24.72%	28.06%	\$0.00
	Aerosol methoprene	84.10%	11.18%	13.33%	15.39%	\$0.71
Totals for all	No Treatment	69.14%	24.32%	27.44%	30.17%	\$0.00
	Aerosol methoprene	83.34%	12.16%	14.20%	16.17%	\$0.71

Table 3-10 Overall Economic Summary of both flour mill and grocery warehouse experiments. The highlighted line represents the treated egg and chemical combination that represent the lowest risk. Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero.

Overall Economic Summary						
Costs: per 10,000 ft ³	3% pyrethrin only	\$ 5.390)			
•	1% pyrethrin plus methoprene	\$ 3.141				
	3% pyrethrin plus methoprene	\$ 5.851				
	Methoprene only plus carrier	\$ 0.711				
Treated Eggs		% Mortality	Risk (90%)	Risk (95%)	Risk (99%)	Cost (\$)
3% Pyrethrin Only	No treatment	57.46%	0.3391	0.3806	0.4164	\$0.00
	Aerosol treatment	80.18%	0.1299	0.1582	0.1902	\$5.39
1% Pyrethrin Plus Methoprene	No treatment	47.31%	0.4382	0.4823	0.5180	\$0.00
	Aerosol treatment	83.06%	0.1082	0.1348	0.1625	\$3.14
3% Pyrethrin plus Methoprene	No treatment	42.76%	0.4821	0.5241	0.5627	\$0.00
	Aerosol treatment	78.39%	0.1492	0.1773	0.2083	\$5.85
Methoprene Only	No treatment	70.17%	0.2391	0.2667	0.2919	\$0.00
	Aerosol treatment	72.06%	0.2182	0.2469	0.2729	\$0.71

CHAPTER 4 - Modification of a Population Growth Model to Simulate Response in Indianmeal moth (*Plodia interpunctella* Hübner) to Methoprene Alone and in Combination With Pyrethrin and Esfenvalerate

Abstract

Management strategies for urban and stored product pests are chosen for a variety of attributes, including cost, efficacy and human safety. As regulations and restriction on the use of insecticides continue to increase, low-impact alternatives to traditional neurotoxins such as insect growth regulators may be valuable management options. As these products become available, there is a need to determine efficacy in regard to survival after exposure, timing of application, and effect on population dynamics, so that overall value of these chemicals can be determined. This study was done to predict the effect of methoprene, and two conventional insecticides combined with methoprene, on survival of eggs and wandering larvae to the adult stage. Effects on populations were simulated using two (wheat-based and raisin diet) modified temperaturebased growth models for the Indianmeal moth. Each insecticide combination was compared at several temperatures under four beginning population levels which differed with diet. Population dynamics varied greatly between the two diets and with increasing temperature. Simulations were also conducted to compare timing and frequency of insecticide treatments for control of the Indianmeal moth, which also were largely different between the two diets. This study also demonstrated that population growth happens so rapidly with this pest that numerous sequential insecticide treatments may be needed to reduce populations, even at temperatures that are not optimal for Indianmeal moth development.

Keywords: temperature, methoprene, Indianmeal moth, development, population dynamics

Introduction

The Indianmeal moth (Plodia interpunctella Hübner) is a cosmopolitan pest of many different commodities, including but not limited to nuts, whole grains, dried fruits, chocolate, beans, flours and meals (Tzanakakis 1959, Simmons et al. 1975, Rees 2004, Mohandas et al. 2007). In addition to the damage caused by Indianmeal moth infestations in raw commodities, it is also a major pest throughout the food manufacturing process (Doud and Phillips 2000, Johnson et al. 2003, Mahroof and Subramanyam 2006). While the Indianmeal moth can be found on numerous food products, development time varies widely with diet (Johnson et al. 1992, Sedlacek 1996). The number of days required for Indianmeal moth development is also influenced by a number of other factors, especially temperature (Howe 1965, Cline 1970), which can heavily influence the number of eggs laid by Indianmeal moth females and the rate of growth in the larval instars (Tzanakakis 1959, Howe 1965, Mohandass et al. 2007). The Indianmeal moth's short life cycle, which can be as short as 19 days (Jenson, unpublished data), combined with the high reproductive capacity of 100-300 eggs per female (USDA, 1986), give it the potential for significant product damage in and around food storage facilities. For these reasons, there is a need to assess how different insecticides and timing of insecticides affect population number and composition.

There has long been an interest in predicting population changes of pest species in response to abiotic and biotic factors. In 1967, one of the first simulation studies was conducted using a computer and a simple model for population development of the red flour beetle (Throne 1995). Since that time, many models have been developed to simulate population growth and decline in response to management practices, especially for stored product pests. Models for the almond moth (*Cadra cautella* (Walker)), the predator *Lyctocoris campestris* (F.), the flat grain beetle (*Cryptolestes pusillus* (Schonherr)), sawtoothed grain beetle (*Oryzaephilis surinamensis* (L.)) and the red flour beetle (*Tribolium castaneum* (Herbst)) have been developed on commodities (Throne 1995). There have also been a number of attempts to modify these models for management tools such as fumigation, biological control and grain protectants (Throne 1995). Many factors influence development of stored product pests; temperature, relative humidity and diet are all important factors to be included in any of these models (Throne 1998). Once a model is developed to predict growth rates on one type of diet, it can be modified to show rates of growth after chemical application or other types of diets (Throne 1995).

The period of time required for Indianmeal moth development from egg to adult is influenced by diet (Johnson et al. 1992), relative humidity (Bell 1975), and temperature (Cline 1970). Temperature can heavily influence number of eggs laid as well as the rate of development in the presence of other satisfactory conditions (Tzanakakis, 1959, Arbogast 2007a, Mohandass et al. 2007). Short exposures to low temperatures (2.4°C) in the egg stage have been shown to decrease survival (Cline 1970), while rearing insects at high temperatures (35°C) has been shown to decrease the ability to reproduce (Johnson et al. 1992). Bell (1975) reported the range of suitable temperatures to be between 15 and 30°C for development and reproduction of the Indianmeal moth. Johnson et al.(1995) demonstrated that the temperature range for Indianmeal moth development was 13-14°C to slightly less than 34°C. There is a wide range of data supporting different biological parameters for Indianmeal moth development (Mohandass 2007). Temperature becomes especially important when our primary insecticide, methoprene, is applied in combination with a chemical that has reduced efficacy with varying temperatures (Arthur et al. 2004). Development is also affected by diet and natural variation of different moth populations. Diet type and quality are also important factors in the developmental time of Indianmeal moth and time from egg to adult can vary substantially with diet type (Mbata and Osuji 1983, Subramanyam and Hagstrum 1993, Johnson et al. 1995).

Jenson (dissertation chapters 1 and 2) has also shown decreased survival to the adult stage when Indianmeal moths are exposed to methoprene, both as a surface treatment and as an aerosol application. These studies (Jenson, dissertation chapters 2 and 3) demonstrated effectiveness of methoprene and methoprene in combination with two conventional insecticides (esfenvalerate and synergized pyrethrin) when insects were directly exposed, as well as when they were exposed to package materials treated with these chemicals. Surface and aerosol treatments of methoprene have different survivorship patterns, as do different insecticide combinations; therefore, predictions of overall population change depend on temperature, commodity type, insecticide type, combination of insecticides, method of insecticide application and timing of application.

The specific objective of this study was to predict response of populations of Indianmeal moth as a result of insecticide applications. For this study, we predicted Indianmeal moth population changes in response to methoprene and to combination treatments with esfenvalerate and synergized pyrethrin at different temperatures by using a computer-based growth model.

This model was developed for Indianmeal moth on whole corn modified for wheat diet and raisins, where one of the primary biological parameters is temperature. We also predicted how the effect of the two different diets would alter survivorship to the adult stage, as well as, timing and frequency of insecticide treatment. Indianmeal moth populations can be substantially reduced with methoprene when used as surface and aerosol treatments, and simulation of that response can be a tool to determine the most efficient timing of methoprene application for control of Indianmeal moth.

Materials and Methods

Model Development for Growth on Wheat Diet

The data set was obtained from Jenson (Chapter 1). A wheat diet was chosen for this study because it represents a complete diet on which survivorship to the adult stage is high (near 100% in species reared in the lab). The enriched diet was composed of cracked wheat and shorts (4.4 kg), brewer's yeast (22g), sorbic and benzoic acid (9.5g each), honey (240ml), glycerin (240ml) and water (120ml). This diet is also representative of a finished stored product that has multiple components (versus a commodity) and some processing or manufacturing. Indianmeal moth eggs used in this study were obtained from a laboratory colony established in June 1988 from individuals collected in Riley County, KS, U.S.A. This colony has been maintained on the same diet described above. The colony from which eggs were obtained were maintained inside environmental growth chambers (Forma-Scientific, Thermo Electron Corporation, Waltham, MA) at 27 ± 1 °C, approximately 50% relative humidity, and darkness (L:D = 0:24 hrs) at the United States Department of Agriculture Agricultural Research Service (USDA-ARS) facility in Manhattan, KS³. Temperature and relative humidity in growth chambers were monitored using digital thermometers and recorded by HOBO data loggers (Onset Computer Corporation, Bourne, MA). Humidity chambers were constructed to ensure consistent relative humidities of 57% using acrylic boxes containing a saturated solution of distilled water and NaBr (Greenspan 1977) and described by Arthur (2000).

³ Kansas State University Museum of Entomological and Prairie Arthropod Research under Lot Number 208.

The model used for these simulations is similar to the *Cadra cautella* (Walker) (Almond moth) model developed by Throne et al. (1998) with components being the time required for the complete life cycle, male longevity, female longevity, and fecundity. Simulations were done using a model for Indianmeal moth originally developed on corn (Throne, unpublished). In the corn model, development time from egg to adult fit a quadratic equation using data from Arbogast's (2007b) study for temperatures 20-35°C, extrapolated to 10-40°C. While there is some evidence that relative humidity and sex affect development rate, they are not included in this equation. For our model to simulate survivorship on wheat diet, data were fit to a linear equation (y=-66.242-1.1513x (R^2 = 0.94)) based on data generated by averaging control treatments for eggs in experiments reported in Jenson (dissertation chapter 1) (Table 4-1). Egg to adult development was fit to a linear equation and extrapolated to 10-35°C. Survivorship did not differ with temperature (it was about 88% across all temperatures); this development is similar to that found by Johnson et al. (1992). Above 32°C, we used a linear regression to predict that no survivorship will occur at temperatures approaching 40°C; below 20°C, survivorship was also set at zero. K values (thermal constants) generated from the Jenson data set were too small for the model because individuals emerged within a 2-3 day time span (insects are very uniform in a carefully controlled colony situation). Therefore, we used K values from the Arbogast (2007b) model.

There are no published data regarding adult male longevity. However, Brower (1976) showed no difference in longevity of males and females at 27°C and at 60% relative humidity. Although not compared statistically, longevity of males and females did not appear to differ at 28°C and 65% relative humidity in a study by Huang and Subramanyam (2003). For the purposes of this model, male longevity data from Arbogast (unpublished) was used so that a linear regression equation was fit from 20-35°C with 75% relative humidity; extrapolated from 10-40°C. Above 40°C, the same longevity information was used as at 40°C; below 10°C, longevity was set to 20 days. For female longevity, the adult females were divided into two groups based on data from Arbogast (unpublished data). Young females were classified as young until they had oviposited 75% of their eggs. Thereafter, they were entered into a group designated as "old" females. Standard deviations needed to calculate K values for longevity were estimated using the method of Shaffer (1983). The model includes no effect of relative humidity on female longevity. Longevity was the same for both groups of females as male longevity at

and above 40°C. For young females below 10°C, longevity was set to 15 days; for old females it was set to 20 days.

Fecundity data in the model were from an unpublished study on fecundity by Arbogast, and data were extrapolated to 15°C based on Mbata's study (1985) on factors affecting oviposition in Indianmeal moth. Effects of relative humidity on fecundity were simulated also based on Mbata's study (1985) using linear regression to reduce fecundity when relative humidity was below 75%. The accuracy of this model's components was checked using a series of manual simulations in lieu of a formal validation on wheat diet. Beginning with one of each life stage, time to emergence or expiration was noted and all components appear to match expected values at temperatures from 5 to 45°C at 5 degree intervals. All simulations run for the purposes of this study were run at one of several constant temperatures with all relative humidities fixed to 57%.

Model Development for Growth on Raisins

Survivorship on raisins was calculated based on means and standard errors of control treatments in Jenson (dissertation, chapter 3). Data for the adjustment of the model for raisins was based on a cumulative mortality and days until adult emergence from several studies conducted to determine effects of aerosol methoprene treatment with Indianmeal moth in various situations, including direct exposure to the insecticide. Eggs used in those experiments were derived from the same colony as described above. Mean survival from experiments in Chapter 3 excluded treated package materials (to avoid interference with development rate) and used treated diet and treated egg controls with raisins as the diet; they were $11.00 \pm 10.27\%$ (approximately 1.7 times the growth rate on the original model for corn.). For development time (number of days) on raisins, from the data in Chapter 3 there are no standard deviations, just a range from 58 to 61 days when treatments were examined and data were collected for total number of emerged adults. Initially, treatments were counted on day 46; then we extended incubation time to approximately 60 days because data from Johnson (1992) showed overall adult emergence was spread out for Indianmeal moth reared on raisins.

Development of the raisin model is more similar to the original model (Throne, unpublished) for Indianmeal moth development on corn than that of the wheat diet model described above. Survivorship was set to 11% from 20-32°C and with no survivorship below

20°C and above 40°C (above 32°C a linear regression was used to predict no survivorship at 40°C, and survivorship at 20°C and below was set to zero). Development time from egg to adult was fit to a quadratic equation (y = 882.6-57.464x +1.0018x², R²= 0.99) extrapolated from 10-35°C, based on the original model's developmental rate adjusted for days to adult emergence observed on raisins (Table 4-2). Adult longevity and fecundity were calculated by the model in the same way as described for the wheat diet. Accuracy of this model's components was checked using a series of manual simulations instead of actual validation data on raisins. Beginning with one egg, one young female, one young male and one old female, time to emergence or expiration was noted and all components appear to match expected values at temperatures from 5-45°C at 5 degree intervals. All simulations in this study were done at one of several constant temperatures with all relative humidities fixed to 57%.

Temperatures simulated for both diets were 21, 24, 27, 30, 32, and 35°C and were chosen to represent a range of temperatures that may be encountered in warehouse, transportation, or food manufacturing facilities. Simulations were run at these temperatures for both diets with different beginning populations. Data points for curves represent total population (adults and immatures) on that day of simulation. It is important to note that aside from the biological parameters placed on the Indianmeal moth populations, and the occurrence of insecticide treatment events, there are no other limiting factors on the populations. This model does not provide for the limitation of population growth based on amount or quality of food source (carrying capacity), space constraints, or multiple diet types present for individuals to access.

Survivorship Data for Exposure to Insecticide Treatments

Because the adjustment for survivorship in the model must be applied to all immatures rather than separate eggs and larval stages, based on the outcomes of previous experiments (Jenson, unpublished, see Chapters 1 through 3) survivorship information for each insecticide treatment is based on data for eggs. Data are available for survival of eggs to the adult stage for all treatment types. Adjustment of survivorship was applied only to immatures because there are no data to support the assertion that methoprene kills adult Lepidoptera. Mortality for each insecticide was calculated using the label rate for surface or aerosol application as this is the most likely scenario for use in field situations.

Mortality was simulated to occur at noon on the day of insecticide treatment for all immatures that would be in that stage at that time. While methoprene extends the larval period in Indianmeal moth, these individuals will most likely not reach the adult stage. Because the goal of this model is to determine effect of treatment combinations on the overall survival of moths to the adult stage, mortality occurring immediately removes these insects from the pool that may become mature and reproduce. Also, although larval data were not used in developing these scenarios, there is evidence that with increasing exposure interval, there is increased mortality for fifth instars; so the possibility that insects may come in contact with the chemical after the treatment interval makes this a reasonable way to account for survivorship. The third factor in determining survivorship for each chemical treatment was that only information for insects reared on wheat diet were used for the wheat model, and insects reared on raisins were used for the raisin model. While insecticide treatments chosen may also reduce adult survival, we have no data to support inclusion of that mortality into the model.

Four insecticide treatments were selected for simulations based on the experiments in Chapters 1, 2 and 3. These were methoprene as a surface treatment, methoprene as an aerosol treatment, methoprene and esfenvalerate applied together as an aerosol, and methoprene and 1% synergized pyrethrins applied together as an aerosol. Because there is no interaction between temperature and methoprene (Jenson, unpublished) the survivorship value of methoprene alone as a surface treatment applied at the label rate is 45.07%. This was calculated based on experiments from Chapter 1 and included survival across all surface types when Indianmeal moth eggs were exposed to the label rate. The survivorship value calculated for methoprene at the label rate delivered as an aerosol was based on data from Chapter 3 in which we had a methoprene-only aerosol treatment. The mean survivorship averaged across all exposure types was 30.99%. Values for the aerosol insecticide treatment combination of methoprene and esfenvalerate were calculated from egg exposure data in Chapter 2, for which both chemicals were delivered at the label rate and were determined to be 17.67% survival. The final insecticide combination used for these simulations was the aerosol treatment of methoprene and 1% synergized pyrethrins (both at the label rate for aerosol application) from data in Chapter 3. The survivorship value of these treatments is 5.37%, the lowest survivorship used in the simulations. Simulations were run at three temperatures (24, 30 and 35°C) for insecticide treatment scenarios, with a beginning population of 100 eggs and no adult moths.

Timing of Insecticide Applications

In addition to mortality, timing and frequency of insecticide applications have a large impact on population levels. We examined three one-time applications of each insecticide (30, 60 and 90 days) from the first day of the simulations (with a beginning population of 100 eggs and no adult moths) at one temperature for wheat diet (24°C) and three temperatures for the raisin diet (24, 30 and 35°C). We also simulated monthly and bi-weekly recurring insecticide treatments for both diets and at three temperatures (24, 30 and 35°C). Monthly and bi-weekly treatments were based on current industry practices.

Results

Development on Wheat Diet and Raisins at Several Temperatures

Simulations of population growth for five scenarios (different beginning numbers of individuals) on the wheat diet for each of six constant temperatures (21, 24, 27, 30, 32, and 35°C) are shown in Figures 4-3 through 4-8. These simulations were run from day 1 to day 180 and used a fixed relative humidity (57%). Scenarios were: beginning with a single egg; one young female; one old female; ten young females; and 10 old females. When ten young females were used, this caused the populations to begin and end at the highest levels compared to the other scenarios. As temperature increased from 21 to 32°C, populations became increasingly larger, while at 35°C populations were the smallest. All scenarios began with slightly different numbers of individuals, but they followed the same trends as populations increased. Results are displayed on a log scale because of rapid population growth, even when simulations were begun using a single individual.

Simulated population development on a raisin diet using four population growth scenarios at six constant temperatures (21, 24, 27, 30, 32, and 35°C) and 57% relative humidity is shown in Figures 4-9 through 4-14. The four population growth scenarios simulated were: beginning with one hundred eggs; one hundred young females; one hundred old females; and one hundred of each young and old females. Simulations were conducted over 180 days and figures were labeled using the log scale for comparison with the wheat diet. Growth curves followed approximately the same trend for all temperatures and, again, 35°C had the lowest rate of population growth.

Response of Populations Following Exposure to Insecticide Treatments with Different Timing and Frequency of Applications

One-time applications of insecticides for wheat diet were simulated beginning with one hundred eggs on day one and run for 180 days on only one temperature (24°C). Using only one temperature was adequate to describe the small effect produced by any of the insecticide treatments (applied only one time). Even the methoprene plus 1% pyrethrin with a survivorship of 5% would result in extensive development of Indianmeal moth populations. Figures 4-15, 4-16, and 4-17 show the overall population effect when each insecticide is applied at 30, 60 and 90 days, respectively, and Figures 4-18 through 4-20 show a more detailed view of population reductions in response to each of these insecticide treatment scenarios.

Figures 4-21 through 4-29 display the effects of one-time applications of insecticide treatments on population development of Indianmeal moths on raisins. Simulations were run beginning with 100 eggs on day one and run for 180 days, with single insecticide applications at 30, 60 and 90 days shown in separate figures. Unlike simulations on the wheat diet, temperatures of 24, 30 and 35°C were used. Single applications at each of these periods at 24 and 30°C greatly reduced populations, but not enough to eliminate the entire population. In contrast, at 35°C the model simulations show a predicted decline in population levels (Figures 4-23, 4-26, and 4-29).

Predicted population responses to monthly and bi-weekly treatments of each insecticide were simulated at 24, 30 and 35°C. While all insecticide treatments reduced total population at the end of 180 days, the populations on wheat showed a continual population increase (Figures 4-30 to 4-32). Predicted population responses were lower on raisins (Figures 4-33 to 4-35). Surface treatments of methoprene, as well as methoprene aerosol treatments and esfenvalerate combination treatments, significantly reduced populations on raisins; but at all three temperatures aerosol combination treatments with 1% pyrethrin done monthly nearly eliminated populations. Bi-weekly treatments with surface and aerosol treatments on wheat followed the same population trend as the monthly treatments (Fig. 4-36 to 4-38), but treatments combinations of methoprene with esfenvalerate and methoprene with pyrethrin significantly reduced populations, and even kept populations to near zero at 35°C (Fig. 4-38). Similarly, for insects on raisins exposed to bi-weekly insecticide treatments, populations were reduced to virtually zero in the first three biweekly treatments (Figures 4-39 to 4-41).

Partial Budget Analysis

Using chemical cost information calculated per 929m² (10,000 ft²) for surface application and 283.7 m³ (10,000 ft³) for aerosol applications, we conducted a partial budget analysis to compare costs of the four insecticide treatments (surface treatment with methoprene, aerosol treatment with methoprene, methoprene plus esfenvalerate, and methoprene plus 1% pyrethrin) with the simulated mortality at each diet (100% survivorship, wheat diet survivorship and raisin survivorship). Chemical and carrier oil costs (for aerosol treatments) were calculated based on current industry prices. Carrier oil costs fluctuate with the global petroleum market, but for the purposes of this analysis they were fixed to \$0.83 per L (\$3.15 per gallon) or \$0.0008 per ml. Economic risk was calculated at three levels (90, 95, and 99%) as deviations below the target goal. In the case of these three experiments, time and equipment cost are fixed and the only variable cost is the cost of the chemical. Risk, in this case, is the inverse of mortality up to the mortality target, at which it is set to zero. Therefore, the downside risk is mortality below the target level. Results are displayed in Table 5-3. Results show that for Indianmeal moth on the raisin diet, a less expensive control strategy such as aerosol methoprene alone may be used for population suppression, compared with the wheat diet where the most aggressive (and expensive) treatment must be used (methoprene plus 1% pyrethrin).

Discussion

Methoprene incorporated into food media has been shown to be highly effective in reducing survival of Indianmeal moth (Strong and Diekman 1973, McGregor and Kramer 1975, Loschiavo 1976, Firstenberg and Silhacek 1976) to the adult stage. Even at very low doses of 2-5 ppm, survival to the adult stage was greatly diminished (McGregor and Kramer 1975, Loschiavo 1976, Fajardo and Morallo-Rejesus 1979). Jenson (unpublished) showed that methoprene can also be an effective management strategy for Indianmeal moth when applied to eggs and wandering larvae directly, both on diet and on packaging materials, as an aerosol. Management is especially effective in combination with conventional insecticides, although simulations show little population decrease in response to these chemicals without the presence of other limiting factors on wheat diet. Survivorship values determining response following insecticide treatments were conservatively calculated to include only mortality to immatures.

This model assumes that adults were not affected by the insecticide treatment, which is another factor leading to higher populations than one might expect in a food warehouse or grain bin.

Indianmeal moth population growth in response to raisin and wheat diets were very different, as would be expected with such a large differences in mean survivorship for each diet. Differences in simulation at different temperatures would also be expected given that survivorship was modeled in wheat to follow a linear regression, while in raisins the model was developed to fit a quadratic equation. The wheat diet is an example of a "worst-case scenario"; it is a product that can support rapid population growth. Populations on the wheat diet were predicted to grow so rapidly that even with an insecticide treatment that produced 94% mortality, population levels rebounded to pre-treatment levels within a few days. Aside from the biological parameters (temperature-dependent development time, survivorship and fecundity) placed on the Indianmeal moth populations and the occurrence of insecticide treatment events, this model does not include other limiting factors such as amount or availability of a food source. This model assumes that all food present is accessible and present in sufficient enough supply to support the extremely high population numbers that we are generating in our simulations. Despite the simulation of these "worst-case scenario" conditions, populations of Indianmeal moth can grow rapidly given their high reproductive capacity. Therefore, management of the Indianmeal moth in any area containing products that contained wheat diet would be crucial to avoid devastating product loss.

With their high reproductive capacity, populations on raisins were also able to reach high levels in less than six months. This is remarkable given that Indianmeal moth typically have low survival and long development times on this commodity (Johnson et al. 1995). Raisins were chosen for this study as a contrast to wheat diet because dried fruit products can be infested by Indianmeal moth and significant damage can occur (Johnson and Vail 1989, Johnson et al. 2002). Also, there is a critical need to replace the fumigant methyl bromide, which has been relied upon heavily for the dried fruit and nut industries (Johnson and Vail 1989, Hilton and Banks 1997, Johnson et al. 2002). Aerosol applications of methoprene, as demonstrated by our simulations, can be a viable alternative, especially when used in combination with conventional insecticides to reduce Indianmeal moth populations on dried fruit.

There are unique challenges to working in a finished stored product or warehouse situation. In a food warehouse or manufacturing facility situation, temperatures typically do not

fluctuate day-to-day, but they do follow broad seasonal patterns. Temperatures during the cooler months may not be sufficient to eliminate populations (Kaliyan et al. 2007a;b, Johnson 2007), especially in climate-controlled facilities that allow population levels to remain high throughout the year. Although this model does not account for movement of individuals in or out of populations, sampling and monitoring can be used to estimate seasonal population fluctuations and pinpoint sources of infestation in food processing and manufacturing situations (Doud and Phillips 2002, Arbogast et al. 2005). There are also constraints on chemicals that may be applied and on the timing of those applications (Arthur 2008), although insecticide treatments are not the only control strategies that can be effective alternative for the use of methyl bromide for management of Indianmeal moth. Heat (Roesli et al. 2003, Mahroof and Subramanyam 2006), biological agents such as granulosis virus (Vail et al. 1991, Bjornstad et al. 1998), and lowtemperature aeration treatments (Mason et al. 1997, Kaliyan et al. 2007b) have all been modeled using similar biological parameters as we used in our simulations. Other management options can be highly effective in controlling this pest, and can be used in commercial facilities such as good sanitation practices (Fields and White 2002) and insect-resistant packaging (Mullen 1994). While results of our simulation study show that contact insecticides or insect growth regulators can be used for population suppression of the Indianmeal moth, frequency of insecticide applications may be more critical than the timing of treatments at which the insecticide is applied. This is especially true when populations are large on a commodity with high survival. Simulations of population dynamics can be an important and useful tool when coupled with other management strategies as part of an overall integrated pest management program. Sampling for insects may be an important part of the management strategy, in that, along with population modeling, may be an early indicator of infestation, so that chemical applications could be made less frequently and in a timely manner.

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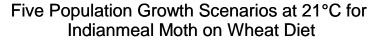
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Figures and Tables

Figure 4-1 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.



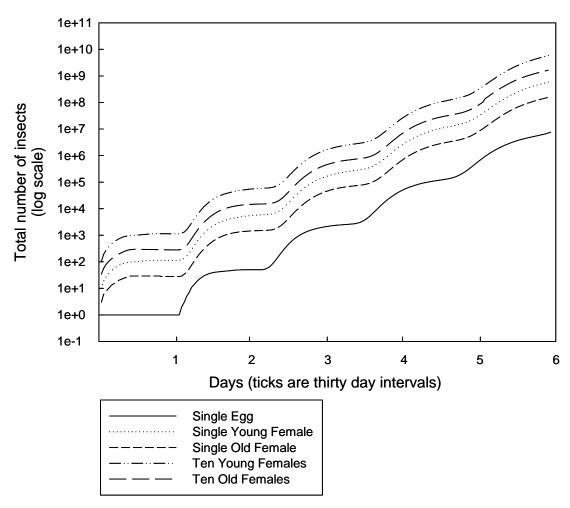


Figure 4-2 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 24°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

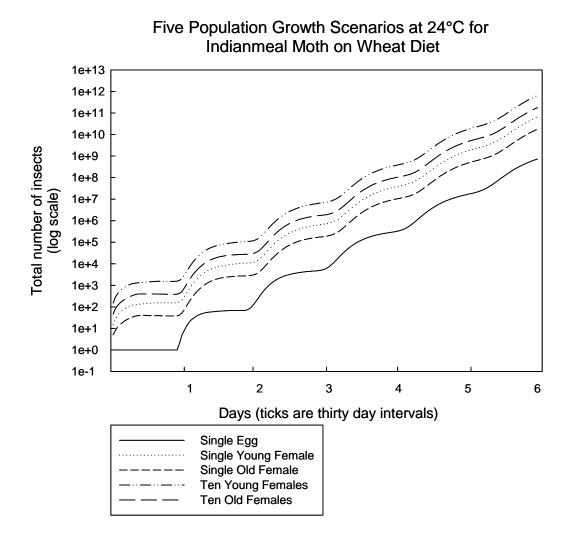


Figure 4-3 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 27°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

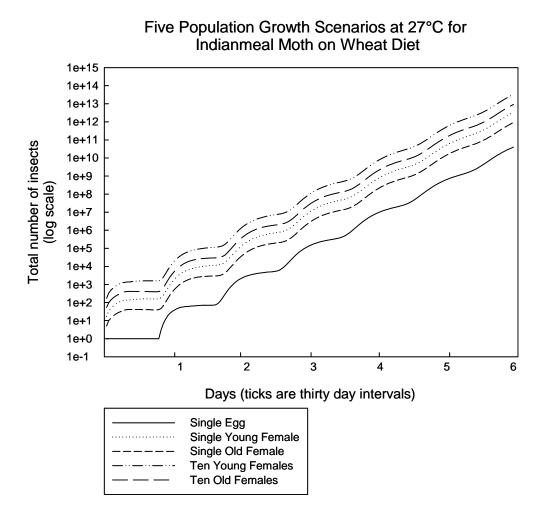


Figure 4-4 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 30°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

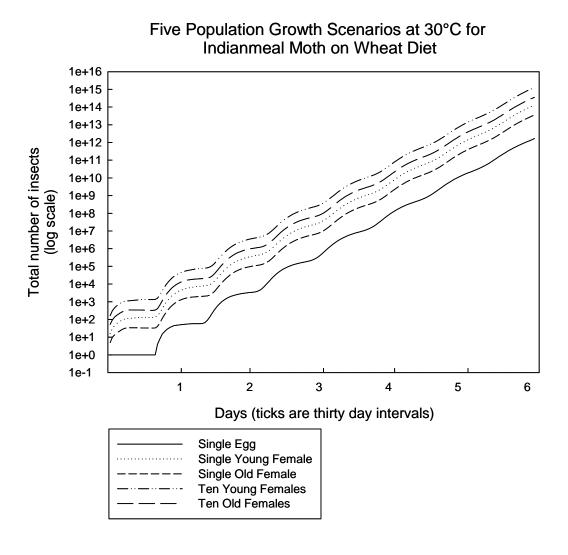


Figure 4-5 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 32°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

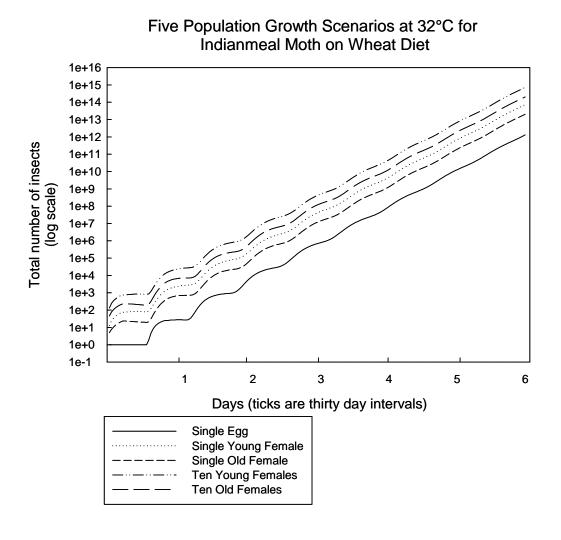


Figure 4-6 Five population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 35°C with 57% relative humidity with the model modified for wheat diet. Scenarios were; beginning with a single egg, one young female, one old female, ten young females and 10 old females. The Y axis is labeled in log scale (common) due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

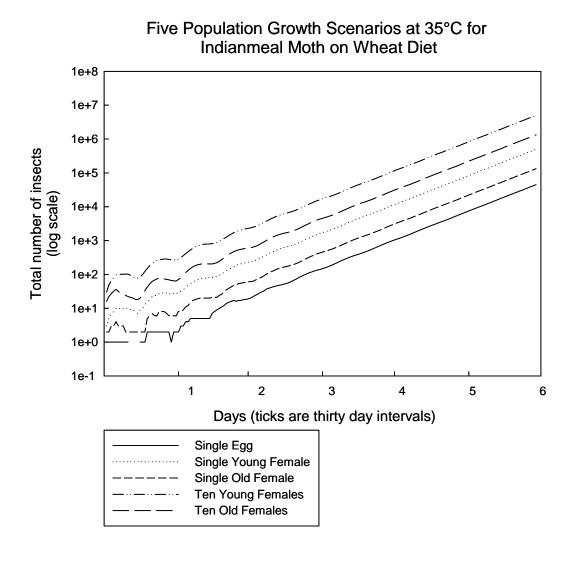


Figure 4-7 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

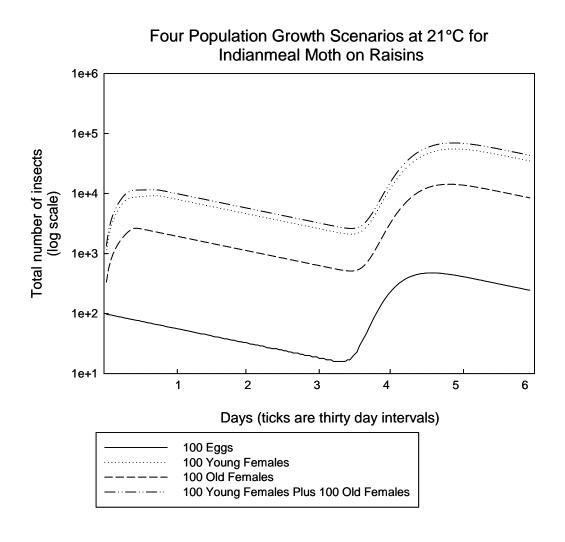


Figure 4-8 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

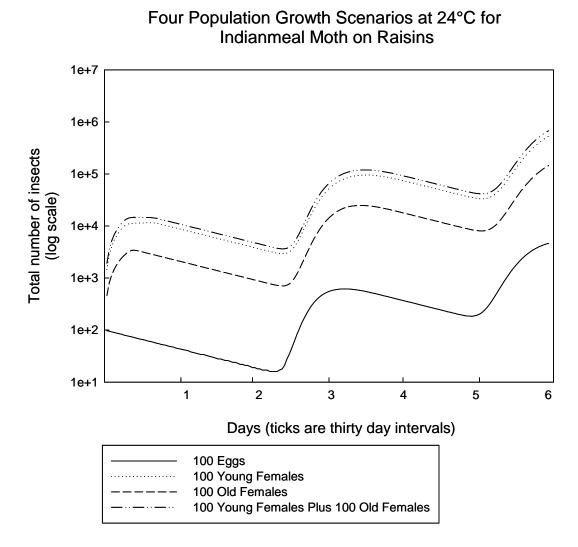


Figure 4-9 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

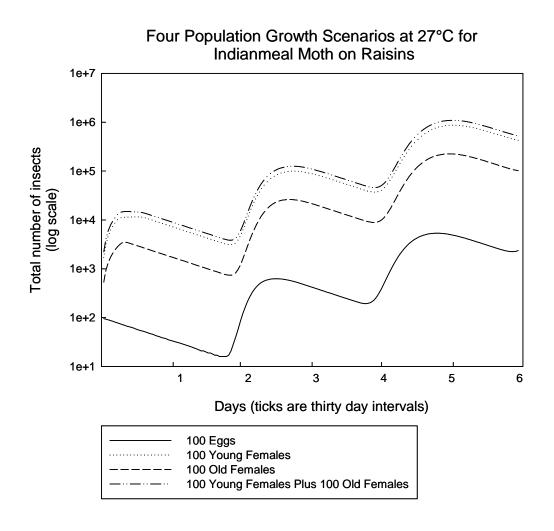


Figure 4-10 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

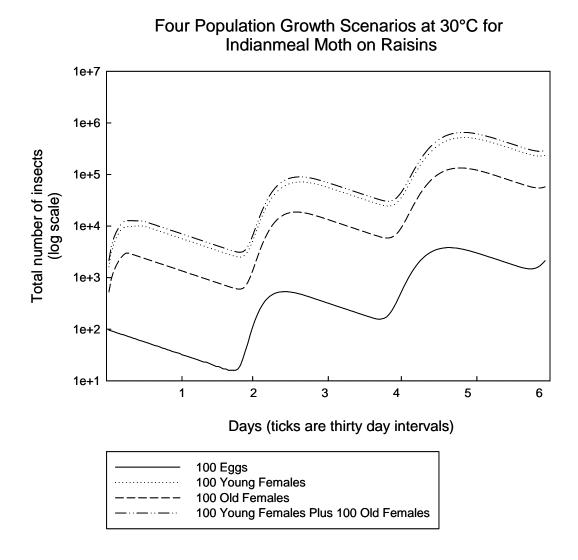


Figure 4-11 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

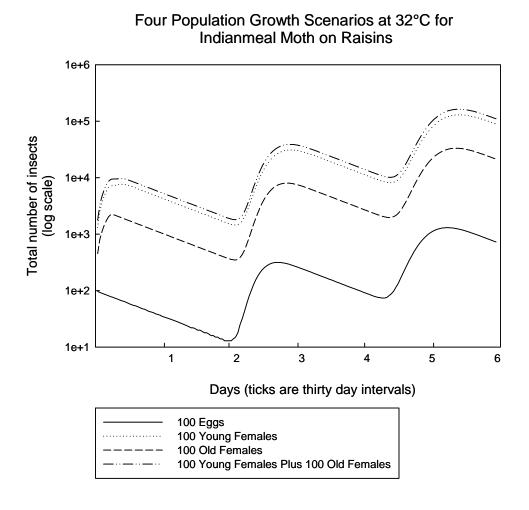


Figure 4-12 Four population growth scenarios (beginning on day 1 of simulation) over 180 days at a constant temperature of 21°C with 57% relative humidity with the model modified for raisins. Scenarios were; beginning with one hundred eggs, one hundred young females, one hundred old females, and one hundred of each young and old females. The Y axis is labeled in log scale (common) for comparison with simulations on the wheat diet and due to the rapid growth at all temperatures. Tick marks on the X axis represent 30 days. Data points are for total population at that day of the simulation.

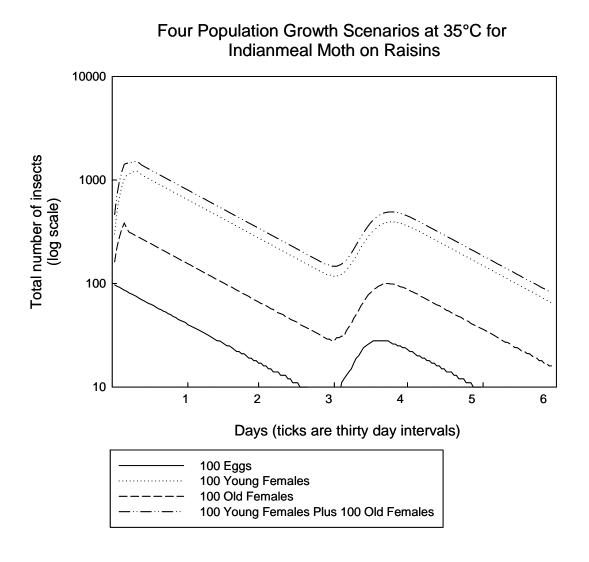


Figure 4-13 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 30.

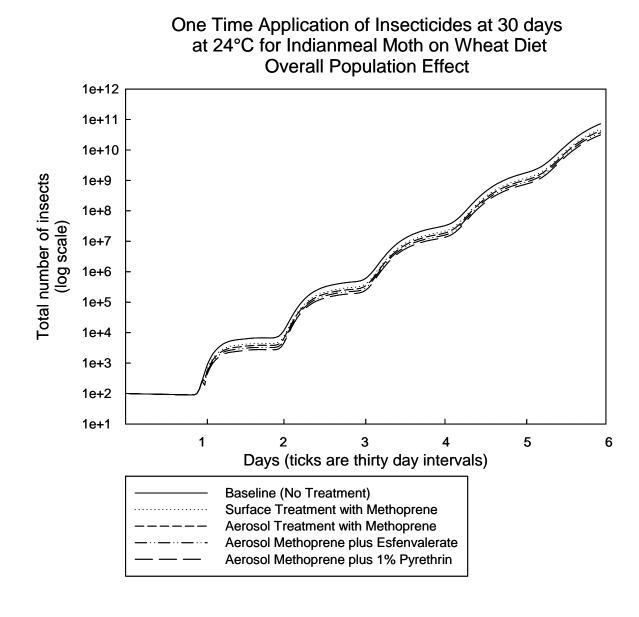


Figure 4-14 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 60.

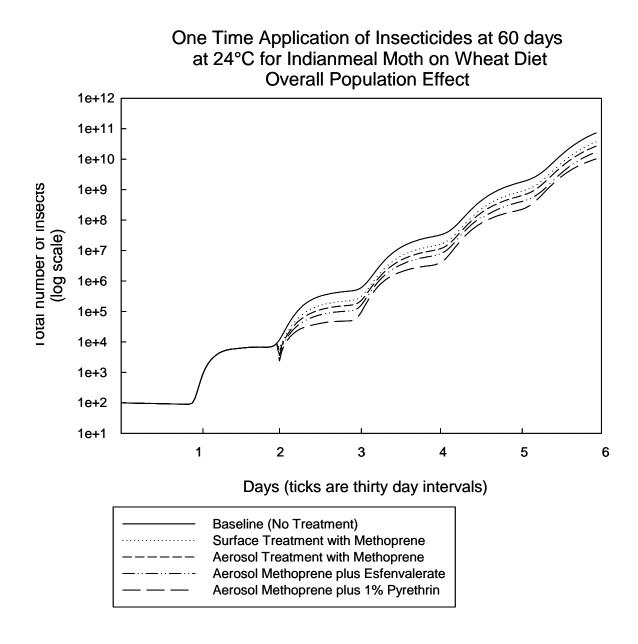


Figure 4-15 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 90.

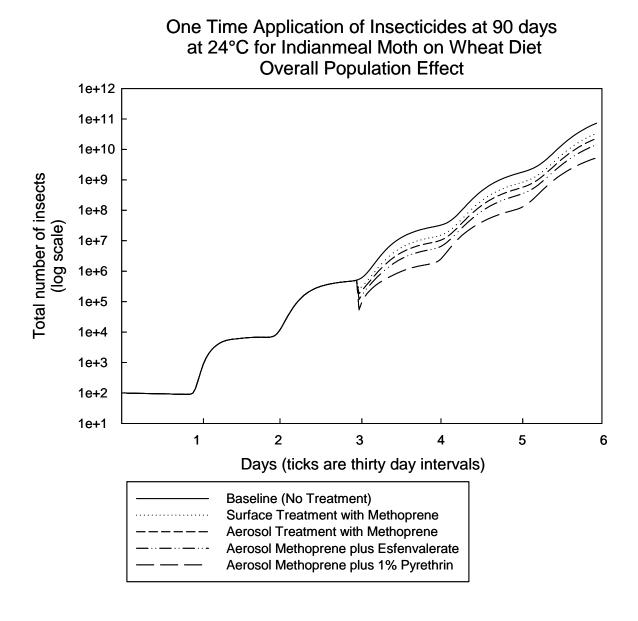
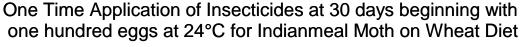


Figure 4-16 A closer examination of the effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 60 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 30.



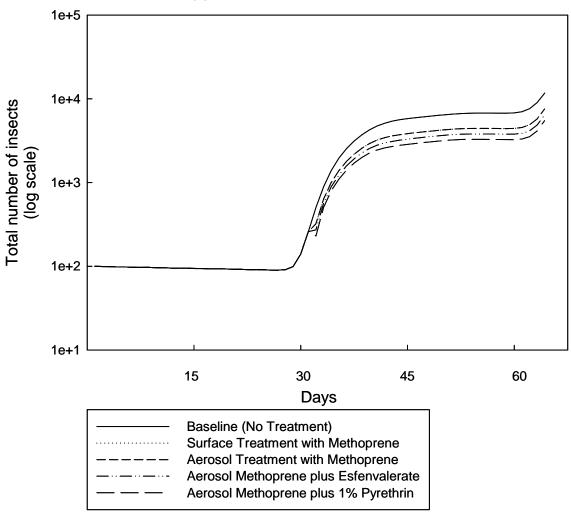
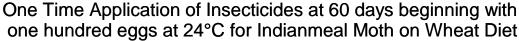


Figure 4-17 A closer examination of the effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 90 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 60.



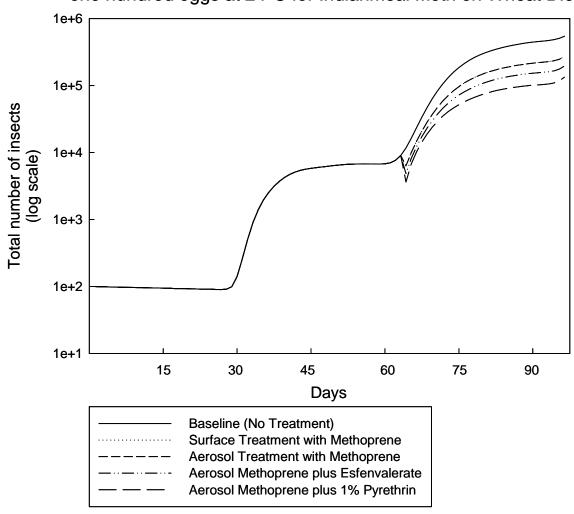
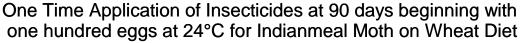


Figure 4-18 A closer examination of the effect of a one-time application of each insecticide treatment on Indianmeal moth living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 120 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 90.



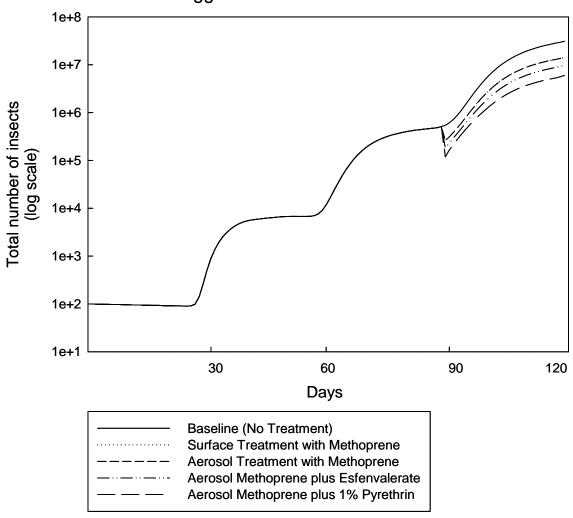


Figure 4-19 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 30.

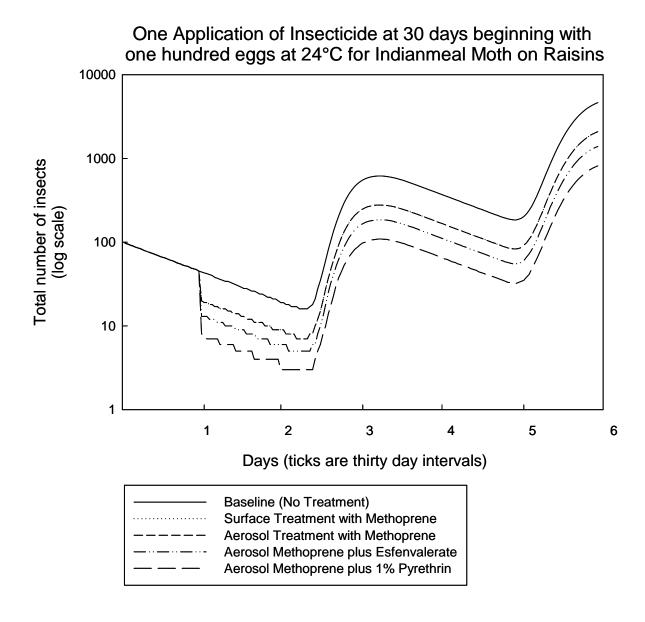


Figure 4-20 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied at day 30.

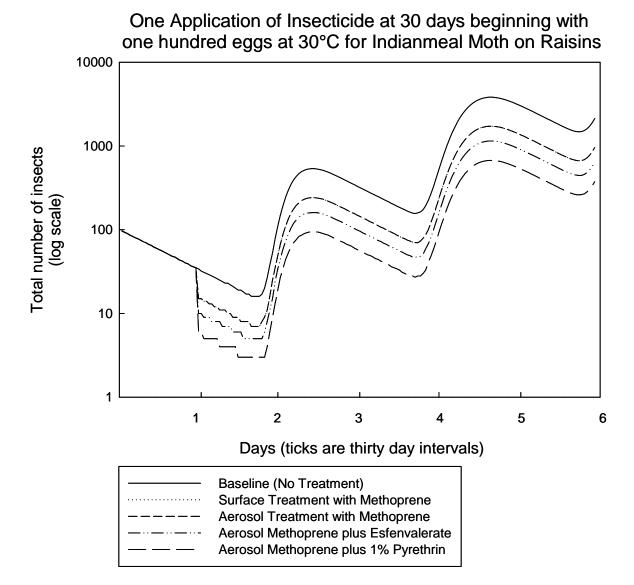


Figure 4-21 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied at day 30.

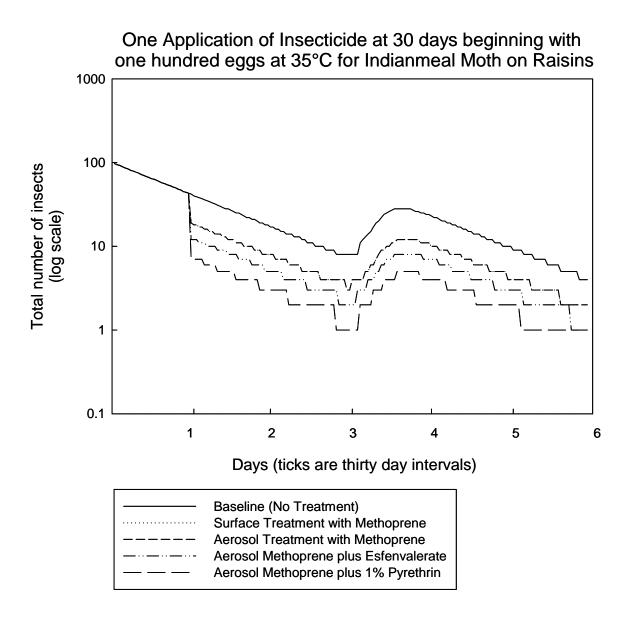


Figure 4-22 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 60.

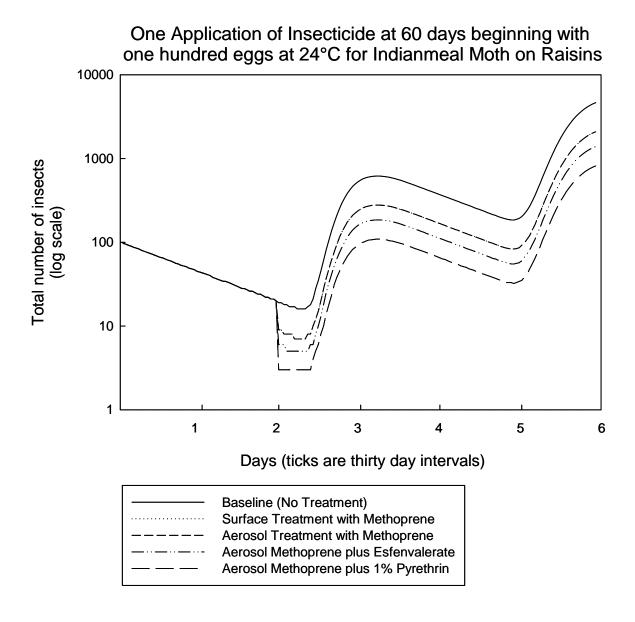


Figure 4-23 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied at day 60.

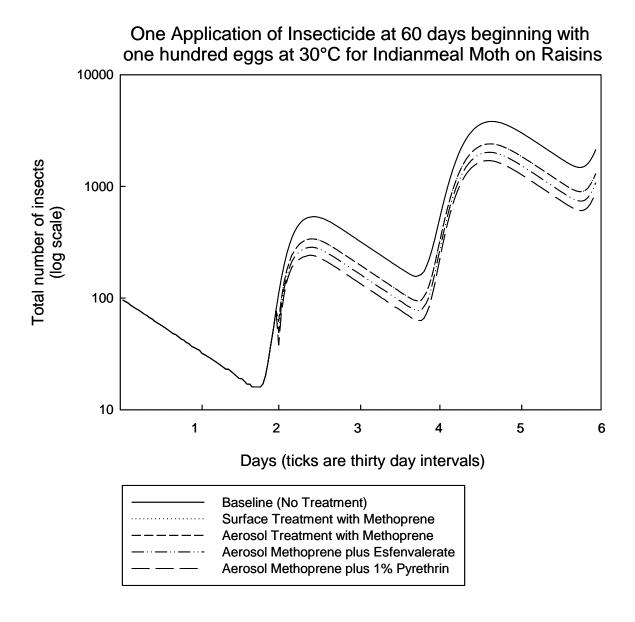


Figure 4-24 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied at day 60.

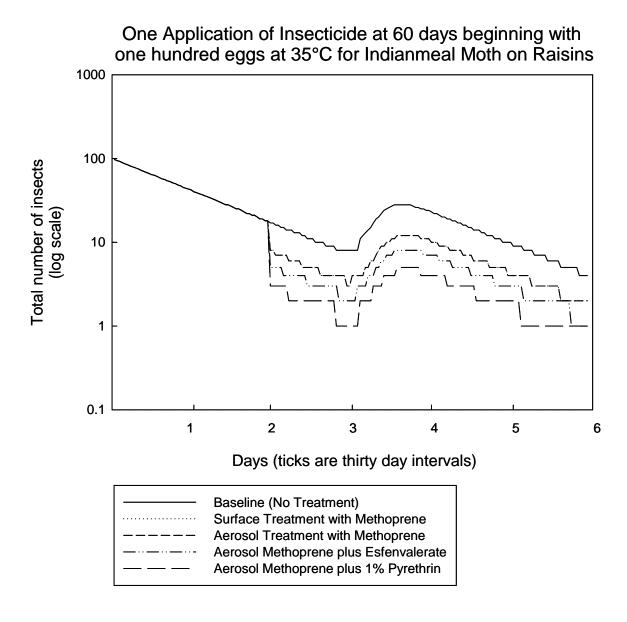


Figure 4-25 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied at day 90.

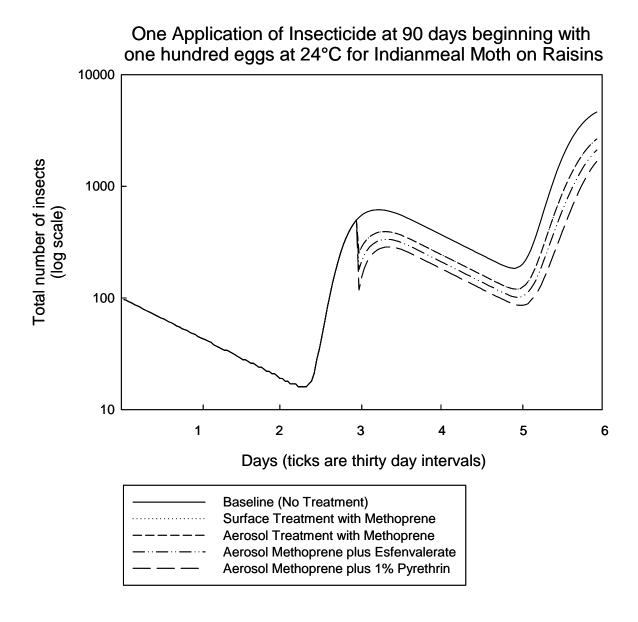


Figure 4-26 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied at day 90.

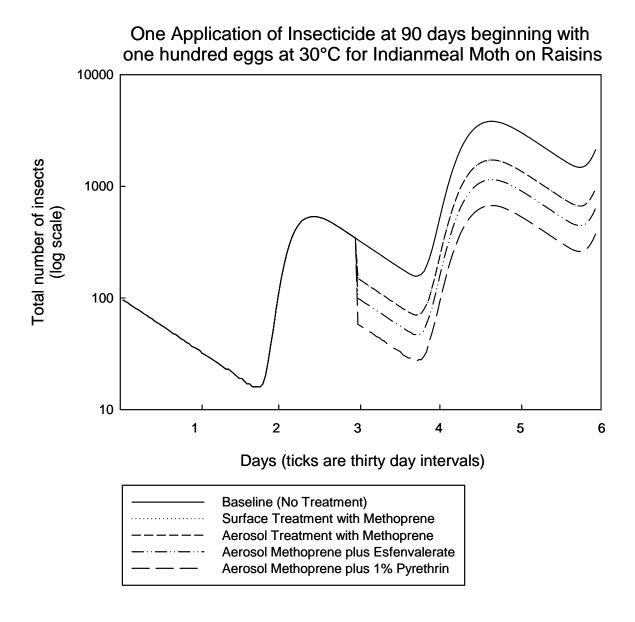


Figure 4-27 The effect of a one-time application of each insecticide treatment on Indianmeal moth living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied at day 90.

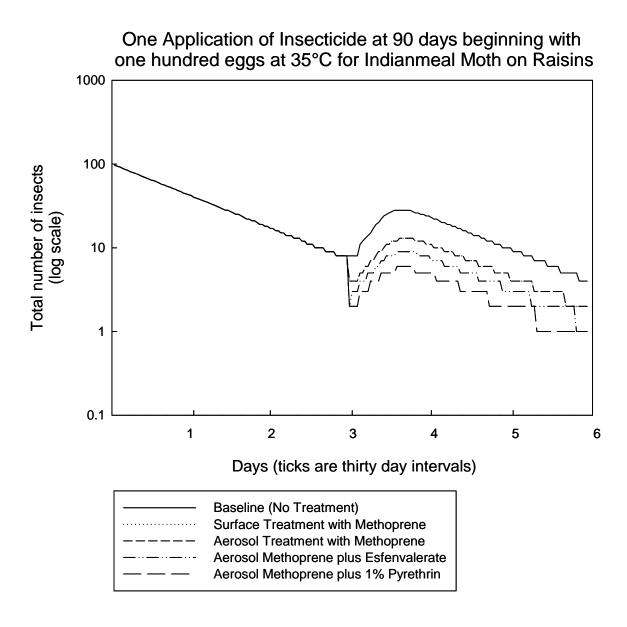


Figure 4-28 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

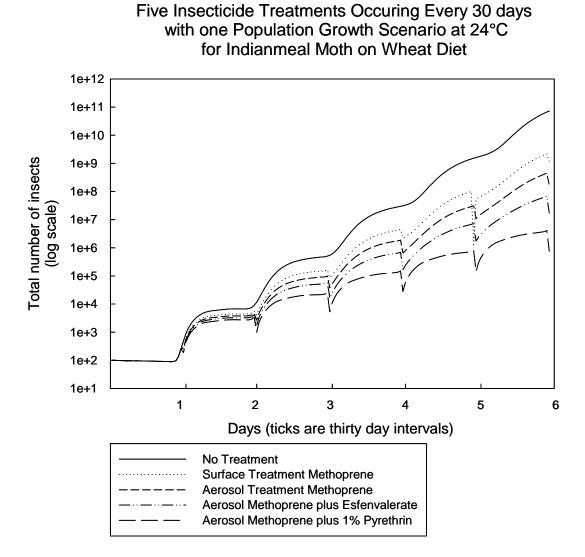


Figure 4-29 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

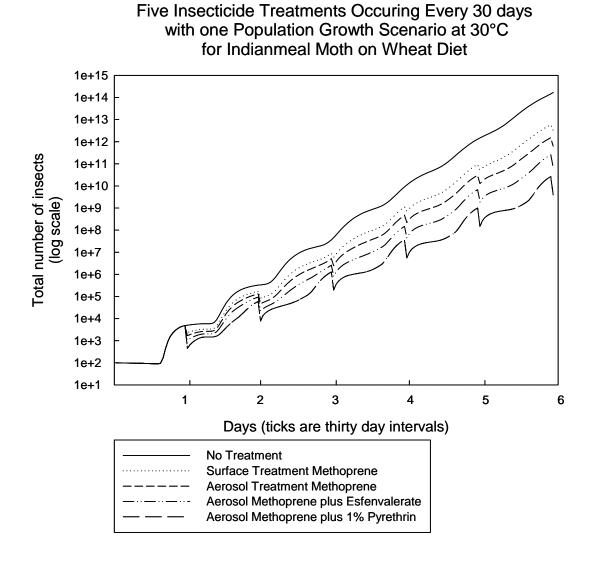


Figure 4-30 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

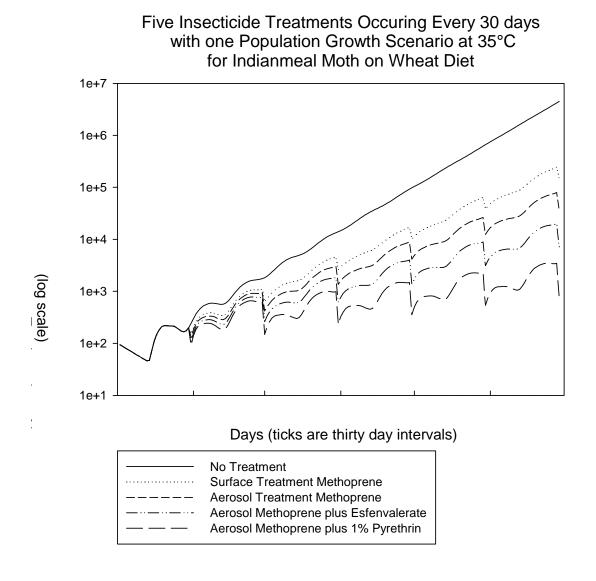


Figure 4-31 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

Five Insecticide Treatments Occuring Every 30 Days with one Population Growth Scenario at 24°C for Indianmeal Moth on Raisins

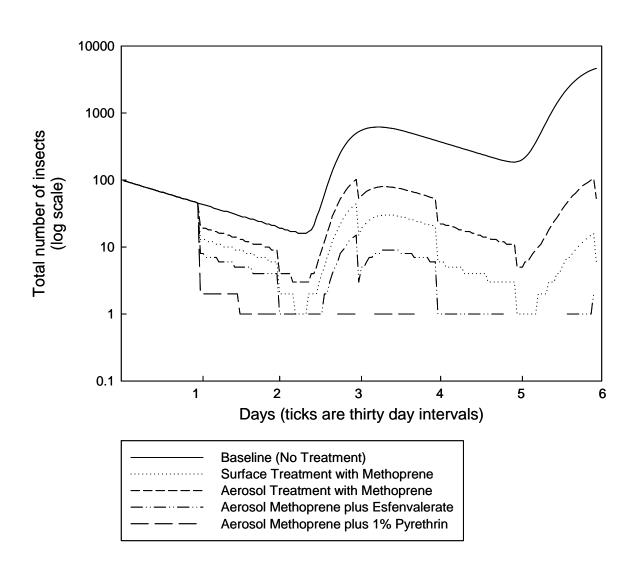


Figure 4-32 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

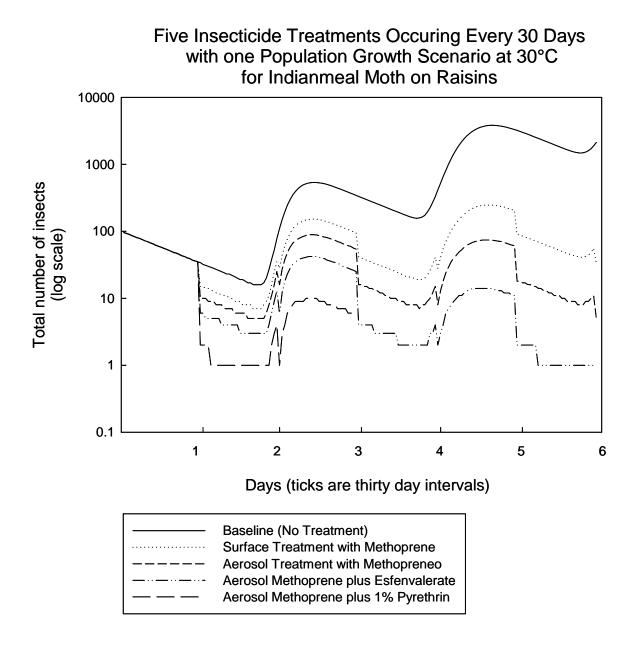


Figure 4-33 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 30 and at 30 day intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

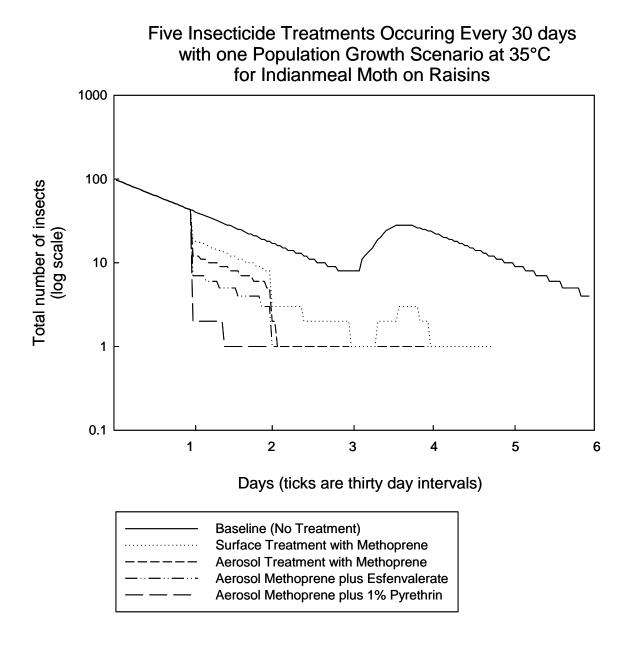


Figure 4-34 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

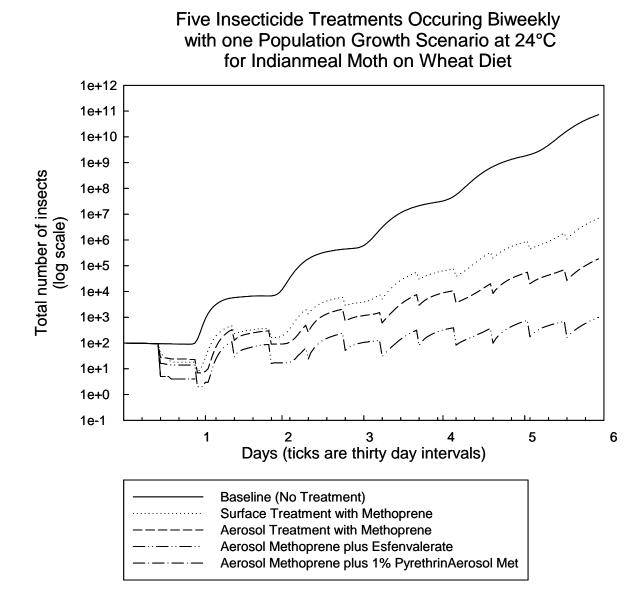
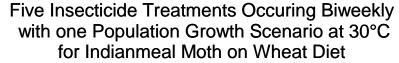


Figure 4-35 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.



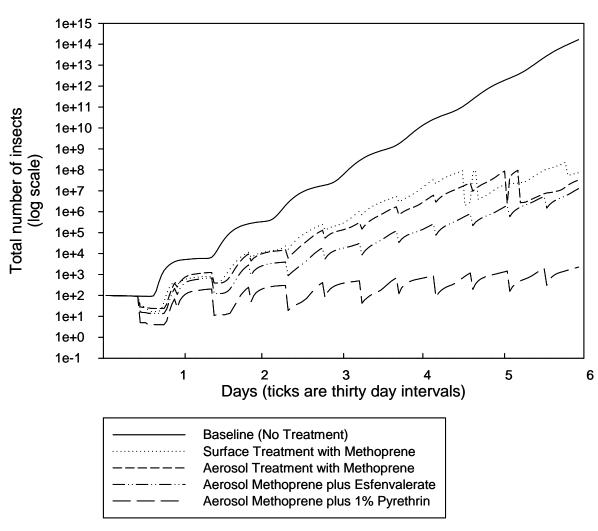
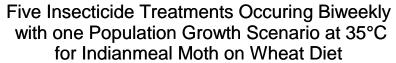


Figure 4-36 The effect of a multiple applications on Indianmeal moth populations living on wheat diet. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.



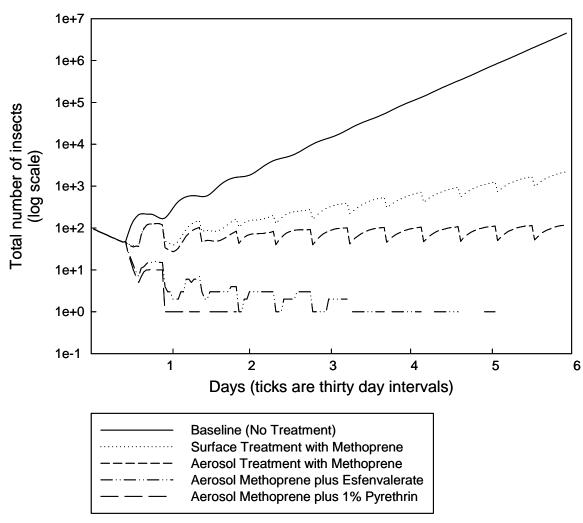


Figure 4-37 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 24°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

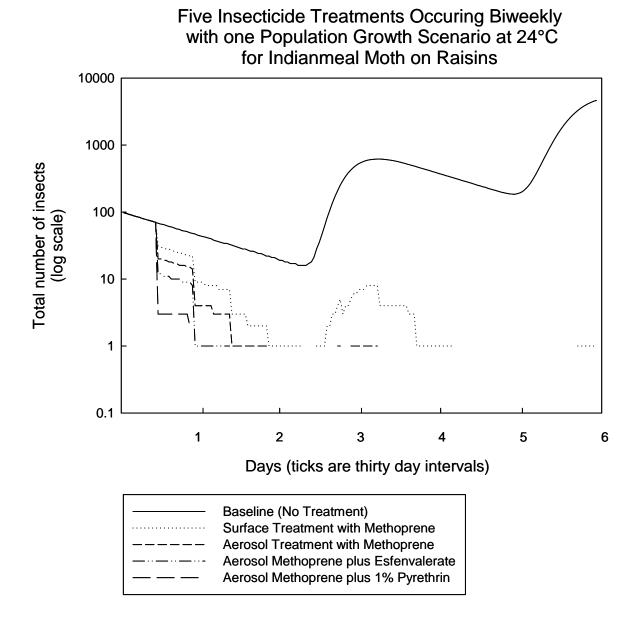


Figure 4-38 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 30°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

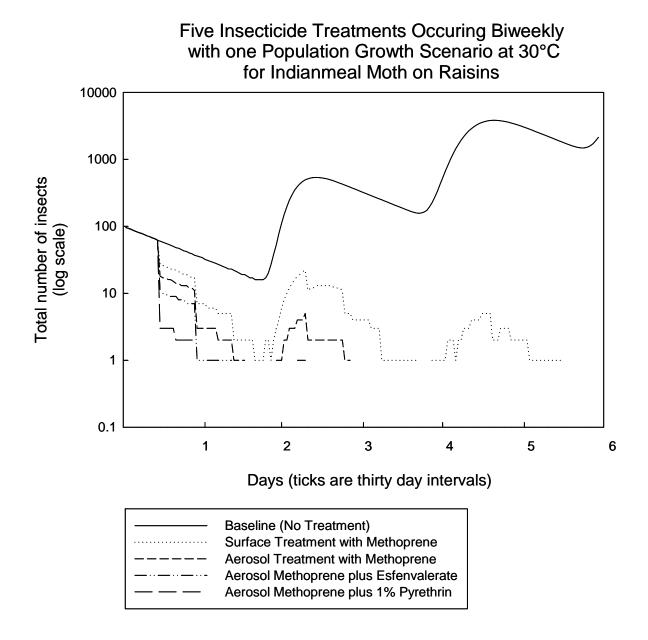


Figure 4-39 The effect of a multiple applications on Indianmeal moth populations living on raisins. One hundred eggs were present beginning on day 1 of simulation and simulations ran over 180 days at a constant temperature of 35°C with 57% relative humidity. Insecticide treatment was applied for the first time at day 14 and at biweekly intervals for the remainder of the simulation. Multiple treatments of each insecticide plus a control are shown.

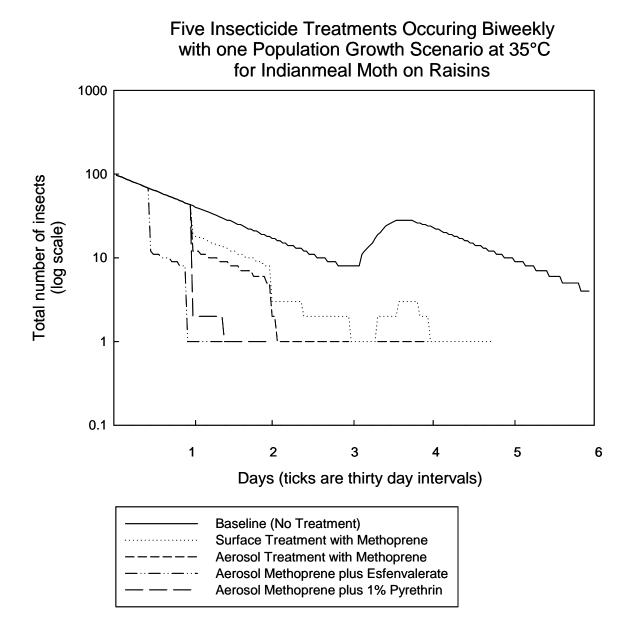


Table 4-1 Survivorship and days to adult emergence by temperature on wheat diet. This information was used to calculate development curve. K values for model were too small due to uniform development in insects used, so values for model were based on Arbogast (2007).

Temp	Level of Treatment	N	Percent Survival		Days to Adu	$K = mean^2/s^{2*}$	
			Mean	SD	Mean	SD	
20	Control	8	87.2819791	11.9079973	36.0833333	11.9503392	9.1
24	Control	12	88.6280463	7.5399454	31.2500000	2.3403574	178.3
28	Control	12	90.0457818	4.8181990	21.0000000	0.8528029	610
32	Control	12	86.9431023	6.4460302	19.3333333	0.7784989	612
mean	•		88.2247274	•	•	•	352.35

Table 4-2 Survivorship and days to adult emergence by temperature on raisins. This information was used to calculate development curve and K values for model.

Temp	Arbogast mean dev. time	Dev. Time on raisins (Arbogast data X 2)	Std. Dev. (Shaffer 1983)	$K = mean^2/s^2$	
20	66.8776062	133.7552124	7.45	322.34	
24	40.7542373	81.5084746	5.19	246.64	
28	29.0347490	58.069498	4.05	205.58	
32	34.9680000	69.936	4.64	227.18	
mean				250.44*	

K < smallest mean/2 DT = 58/(2*(1/24)) = 58 * 12 = 696; use k = 250

Table 4-3 Summary of costs and risk levels for comparison of four insecticide treatments methoprene alone at the label rate for surface and aerosol, methoprene combined with esfenvalerate at the label rate, and methoprene combined with 1% synergized pyrethrin at the label rate) for insects on wheat diet and raisins. Three mortality scenarios are presented; 100% survivorship, 88% survivorship (wheat diet) and 11% survivorship (raisins) (assuming temperature at 20-32°C). Risk is presented as three thresholds; 90, 95 and 99% mortality above which risk is set equal to zero. Costs reflect insecticide and carrier oil costs as explained above. The scenario that represents the lowest risk at any threshold is the methoprene in combination with the 1% pyrethroid treatment, this treatment also represents the highest cost.

Methoprene- Surface		\$ 1.56			
Methoprene- Aerosol		\$ 0.711			
Methoprene plus Esfenvalerate		\$ 1.17			
Methoprene plus 1	% pyrethrin	\$ 3.141			
% Mortality	Risk (90%)	Risk (95%)	Risk (99%)	Cost (\$)	
0.00	0.90	0.95	0.99	0.00	
0.55	0.35	0.4	0.44	\$1.56	
0.6901	0.2099	0.2599	0.2999	0.711	
0.8233	0.0767	0.1267	0.1667	1.169	
0.9463	0.0000	0.0037	0.0437	3.141	
_					
0.12	0.78	0.83	0.87	0.00	
0.516	0.384	0.434	0.474	\$1.56	
0.7273	0.1727	0.2227	0.2627	0.711	
0.8445	0.0555	0.1055	0.1455	1.169	
0.9527_	0.0000	0.0000	0.0373	3.141	
	Methoprene plus I Methoprene plus I % Mortality 0.00 0.55 0.6901 0.8233 0.9463 0.12 0.516 0.7273 0.8445	Methoprene- Aerosol Methoprene plus Esfenvalerate Methoprene plus 1% pyrethrin % Mortality Risk (90%) 0.00 0.90 0.55 0.35 0.6901 0.2099 0.8233 0.0767 0.9463 0.0000 0.12 0.78 0.516 0.384 0.7273 0.1727 0.8445 0.0555	Methoprene- Aerosol \$ 0.711 Methoprene plus 1% pyrethrin \$ 3.141 % Mortality Risk (90%) Risk (95%) 0.00 0.90 0.95 0.55 0.35 0.4 0.6901 0.2099 0.2599 0.8233 0.0767 0.1267 0.9463 0.0000 0.0037 0.12 0.78 0.83 0.516 0.384 0.434 0.7273 0.1727 0.2227 0.8445 0.0555 0.1055	Methoprene Aerosol \$ 0.711 Methoprene plus Esfenvalerate \$ 1.17 Methoprene plus 1% pyrethrin \$ 3.141 % Mortality Risk (90%) Risk (95%) Risk (99%) 0.00 0.90 0.95 0.99 0.55 0.35 0.4 0.44 0.6901 0.2099 0.2599 0.2999 0.8233 0.0767 0.1267 0.1667 0.9463 0.0000 0.0037 0.0437 0.516 0.384 0.434 0.474 0.7273 0.1727 0.2227 0.2627 0.8445 0.0555 0.1055 0.1055	

11% Survival Simulating Raisins as Diet					
No treatment	0.89	0.01	0.06	0.1	0.00
Methoprene- Surface		0.9395	0	0.0105	\$1.56
Methoprene- Aerosol	0.9659	0.0000	0.0000	0.0241	0.711
Methoprene plus Esfenvalerate	0.9806	0.0000	0.0000	0.0094	1.169
1% Pyrethrin	0.9941	0.0000	0.0000	0.0000	3.141

CHAPTER 5 - Economic Feasibility of Methoprene Applied as a Surface Treatment and as an Aerosol Alone and in Combination with Two Other Insecticides

Abstract

Economic evaluations of integrated pest management options are becoming increasingly important as restrictions on conventional insecticides continue to become more stringent and costs of chemical control strategies rise. Aerosol treatments with insect growth regulators alone, and in combination with conventional contact insecticides, can be a feasible alternative to expensive and dangerous fumigants such as methyl bromide for control of the Indianmeal moth (*Plodia interpunctella* Hübner). Mortality of Indianmeal moth eggs exposed to surface-applied methoprene, aerosol methoprene alone, and in combination with esfenvalerate and synergized pyrethrins was 55, 69.01, 82.33 and 94.63%, respectively. The effect of temperature on development time makes frequency and timing of insecticide applications very important as evidenced by simulations of population levels in response to a variety of treatment dates by diet. It also becomes critical in situations where survival of Indianmeal moth is high. Using a measurement of risk that is equal to deviations below a target mortality goal (99%), we were able to optimize the cost and frequency of application using simulated mortality data for each of the treatment strategies. Optimal timing of each insecticide treatment depends heavily on the rate of development based on diet.

Keywords: methoprene, esfenvalerate, synergized pyrethrin, Indianmeal moth, economic analysis

Introduction

Integrated pest management in stored products relies on a variety of management tools for control of pest insects. One traditional insect management practice in mills and dried fruit and nut warehouses (Johnson and Vail 1989, Johnson et al. 2002, Arthur 2008) is to fumigate with methyl bromide. However, this product is scheduled to be phased out under a world-wide agreement, the Montreal Protocol (Fields and White 2002, Anonymous 2004). Some industry groups involved in food production and transportation for human consumption have received a critical use exemption (CUE) for the continued use of methyl bromide (Arthur 2008); but replacement strategies will soon need to be employed (Dowdy 2002).

One current, largely unexplored management option is aerosol applications of conventional insecticides and insect growth regulators (IGRs) to control insect pests in food storage and manufacturing facilities. Aerosol space applications can be an effective way to treat the interior surfaces and storage areas of warehouses and food processing facilities, as evidenced by recent studies (Arthur and Campbell 2007, Arthur 2008) demonstrating aerosol pyrethrin for management of the red flour beetle, Tribolium castaneum (Herbst). Systems for ultra low volume (ULV) aerosol delivery have been designed for and installed in commercial milling and storage facilities. Currently, in facilities where aerosol fogging systems are installed, pest managers are using conventional insecticides alone, and in combination with insect growth regulators (IGRs). IGRs are insecticides that mimic various hormones involved in the developmental processes in insects. They have been proposed for the control of stored-product insects (Oberlander et al. 1997, Mondall and Parween 2000, Campbell et al. 2004, Mohandass et al. 2006a). One IGR, methoprene, which is a juvenile hormone analog, has been evaluated alone and in combination with esfenvalerate and 1% synergized pyrethrin for control of eggs of the Indianmeal moth (Plodia interpunctella Hübner) (Jenson, Chapters 1-3). Comparisons can also be made between the use of methoprene as an aerosol and as a contact surface treatment an in food storage and processing facilities.

Economic analyses of other methyl bromide alternatives using enterprise budgets for specific field crops have been developed (Nelson 1996, Byrd et al. 2006). Partial budget analysis comparing costs of control strategies are used to determine levels of risk associated with each strategy (Boehlje 1984). Using cost information from our partial budget analysis (Jenson Chapters 1-4), along with a population growth model simulating development in response to multiple diet types (Jenson, Chapter 4), a modified economic analysis could allow food

production plant managers and warehouse managers to make decisions regarding control of a single pest species by comparing costs, efficacy of different treatments, and frequency of application of those treatments. Using population growth models to simulate consequences of management decisions can provide even more insight. With extremely low thresholds in finished stored product situations, a slightly different approach from traditional economic injury levels is needed (Higley and Wintersteen 1992, Stejskal 2002; 2003). Many types of economic analysis have already been applied to other systems, including field crops and ornamentals (Headley and Hoy 1987, Jetter et al. 1997) and grain bins (Tilley et al. 2007). However, the warehouse is a novel environment for applying standard methodologies. The objective was to show how these types of analyses may be optimized for insecticide applications in warehouse environments. The specific hypothesis was that there will be significant differences in the cost-benefit structure for each of the insecticide treatment scenarios. The specific goals were to: 1) ascertain how to best utilize the three aerosol insecticide treatments by optimizing timing and frequency of applications and 2) examine the effects of a different food source in response to insecticide applications.

Materials and Methods

Computer Simulations of Mortality due to Various Scenarios of Temperature, Timing and Frequency of Insecticide Application

Simulations of the effect of timing and frequency of insecticide treatments were conducted using a population growth model modified for wheat and raisin diets (Jenson, Chapter 4). This model was based on four main components: time required for the complete life cycle, male longevity, female longevity, and fecundity. Survivorship values for this model were calculated from data for mortality when insecticides were applied to eggs (Jenson, Chapter 1). Adjustment of survivorship was only applied to immatures because there are no data to support the assertion that methoprene kills adult Lepidoptera. Mortality was simulated to occur at noon on the day of insecticide treatment for all immatures that were in that stage at that time. Survivorship values were calculated using the mortality data for Indianmeal moth eggs exposed to the label rates for surface or aerosol application of each insecticide reared on the corresponding diet (wheat diet or raisins)(Jenson, Chapters 1-3), as this is the most likely scenario for use in field situations.

The survivorship value calculated for methoprene at the label rate delivered as an aerosol was based on data from Chapter 3, which evaluated a methoprene-only aerosol treatment. The mean survivorship averaged across all exposure types was 31.0%. Values for the aerosol insecticide treatment combination of methoprene and esfenvalerate were calculated from egg exposure data in Chapter 2, where both chemicals were delivered at the label rate and survival was estimated as 17.7%. The final insecticide combination that was used for these simulations was the aerosol treatment of methoprene and 1% synergized pyrethrins (both at the label rate for aerosol application) from data in Chapter 3. Survival was estimated as 5.4%, which was the lowest survivorship used in the simulations. Although there is no interaction or relationship between temperature and methoprene in survival or mortality of Indianmeal moth between 20 and 32°C (Jenson, unpublished), there is a strong relationship between temperature and total development time from egg to adult. Beginning populations were standardized to 100 eggs and no adult moths. Simulations were run at six temperatures (21, 24, 27, 30, 32, and 35°C) for various insecticide treatment scenarios. Simulation of different temperatures was necessary to evaluate the specific timing of insecticide applications because of the proportion of immatures present in the population on the day of treatment.

Frequency and timing of treatments scenarios were chosen to represent various industry practices. In addition to mortality from treatment and response of population to temperature, timing and frequency of insecticide applications had a large impact on population levels (Jenson, Chapter 4).

Calculation of Costs

For the purposes of this economic analysis, shut-down time and equipment costs were fixed and the only variable costs were the costs of the chemical, the carrier for the applications, and the combinations of carrier and insecticides. Costs associated with methoprene surface treatments were calculated using current industry costs for Diacon II®, calculated per 929.03 m² (10,000 ft²) at the label rate for surface applications (Central Sciences 2002, 1 ml/94m²). Costs for aerosol methoprene treatments were calculated in the same way for per 283.7 m³ at the label rate for aerosol space treatments (900 mg of active ingredient per 10,000 ft³) plus the cost of oil carrier. The costs of the oil carrier were fixed for the purposes of this analysis as to \$0.83 per L (\$3.15 per gallon) or \$0.0008 per ml. However, the cost of oil carriers may fluctuate with the

global petroleum market. Current prices for esfenvalerate (Conquer®) and 1% synergized pyrethrin (Entech Fog-10®) were calculated based on their labeled rates for aerosol delivery systems (Paragon Products, Entech) per 283.7 m³. Costs for combination treatments were calculated by adding together the specific insecticide costs and adjusting the cost for carrier oil.

Target Mortality Model and Optimization

Tilley (2007) modified a target minimization of total absolute deviations (MOTAD) model (Tauer 1983) for grain infestation into a model for modeling risk-and-return in heat disinfestations of grain bins. This empirical model (Target MOTAD) is useful in analyzing trade-offs between risk and return to maximize return above a critical limit (Tauer 1983). Risk and return are directly related; therefore decreasing returns are associated with decreased risk levels (Tilley et al. 2007). Return in this study is defined as mortality of the target insects as a result of a specific management intervention; therefore, the target return was set to a threshold of 99%. The threshold for this model was set at such a high level to provide a realistic threshold for infestation food processed and stored for human consumption (which is virtually zero insects per unit). Risk, in this case, is the inverse of mortality up to the target mortality target of 99%. Cost structure is calculated using the price of the chemical as the relevant cost. Labor and equipment costs are fixed between treatments. Modification of Tilley's model for optimization of cost, risk, and frequency of insecticide treatment allowed us to minimize risk and maximize mortality. The tradeoff in this case is lowered cost and increased risk for treatments that allow for higher survival of the pest insect.

Mortality indexes were obtained by simulating mortality of Indianmeal moth eggs exposed to six scenarios: no treatment, one treatment at day 28, and recurring treatments every 2, 3, 4, and 6 weeks beginning at day 28. These treatments were simulated across six temperatures (21, 24, 27, 30, 32, and 35°C) separately for both a wheat and raisin diet. Survivorship estimates for insecticide treatments were modeled based on results from studies by Jenson (unpublished, Chapters 1-3) and calculated for use in population simulations in Chapter 4 and in this study. Cells for mortality levels in the economic model were calculated by subtracting the population total at 180 days for each treatment from the total population surviving at 180 days where there was no treatment. Therefore, mortality indexes for the target mortality model are the percentage

reduction in population size from the baseline population at each temperature. Cost was minimized based on the allowable deviations below the target 99% mortality rate.

Results

Costs of each treatment and cumulative costs for each treatment scenario over the 6 month interval are displayed in Table 5-1. Costs ranged from \$0.71 to \$3.14 per treatment and from \$7.82 to \$34.55 for 6 months of biweekly treatments per 10,000 ft³ of facility headspace, which caused the individual costs of specific treatments to be significantly different. Mortality levels (explained above) were not correlated with treatment cost (i.e., the least expensive treatment did not always have the lowest cost). Population levels are presented in Tables 5-2 and 5-3. Inclusion of these data were necessary because optimization of the economic model showed that there was a 99% or more reduction in some of the treatment scenarios. However, due to unequal survivorship on wheat and raisins (88% and 11%, respectively) based on data from Jenson, Chapter 4), there were often considerably more surviving individuals on wheat diet, although insecticide treatments reduced survival proportionally for both diets.

Tables 5-4 and 5-5 show the results of the model fit to the methoprene plus 1% synergized pyrethrin treatments. Minimization of costs at risk level 1 (the lowest realistic amount of risk) was \$5.98 per treated unit, and the optimal mix of treatments was between our one-time treatment at day 28 and treating every 6 weeks starting from day 28 in an environment where both diets were present beginning with 100 Indianmeal moth eggs on day 1. Lower cost and higher risk alternatives are displayed in Table 5-5 as well. The insecticide treatment option of methoprene plus esfenvalerate applied as an aerosol is modeled in Table 5-6, with solutions in Table 5-7. The optimal mix of timing scenarios at risk level 1 was to treat with these insecticides at an interval between 3- and 4-weeks (following an initial treatment at day 28) at the cost of \$3.63 per unit area. The risk level 0 scenario, is not realistic, and even at \$9.24 per treatment this scenario cannot be optimized. Tables 5-8 and 5-9 present the target mortality model for aerosol methoprene treatments. Overall these treatments are the least expensive option, but have a mean of only 59% mortality per treatment. However, looking at the optimal mix of treatments, treating with aerosol methoprene between 3 and 6 weeks can cost as little as \$2.40 per unit area and keep risks minimized near the 1 level. Surface application of methoprene can be optimized for costs

when treating one time at 28 days and six weeks thereafter for \$5.87 per treatment unit (Tables 5-10 and 5-11).

Discussion

Treatments in our economic model are based on a mix of two diets (wheat and raisins), on which Indianmeal moth survives very well and very poorly, respectively. These types of simulations are realistic given that warehouse environments may have many different food types stored in close proximity to one another. Possibilities for simulations are limited only by available pest population growth models and variable cost information. While our simulations and economic models do not include every possible treatment scenario, we have presented a range at which decisions about insecticide type, application type, and frequency could be made under our model conditions. It may be possible to compare and optimize for non-chemical pest management options such as chilled air aeration, ambient air aeration, fumigation and heat treatments of facilities (Mason et al. 1997, Maier et al. 1997, Rulon et al. 1999, Tilley et al. 2007), which may be used to control the Indianmeal moth and other stored product insects. Our results are especially useful when comparing management strategies, such as those listed, with costs of low risk insecticides.

The warehouse environment is a new and novel environment for economic simulation. Integrated pest management practices have long employed the concepts of economic and aesthetic injury levels in field crop systems and in biological control programs (Stejskal 2002, 2003). Integrated pest management in a finished stored products situation presents unique opportunities and challenges because of the low tolerance for insects and the tangible and intangible costs associated with insect infestation. Integrated pest management in field crops has a successful history of using economic injury levels and economic thresholds to determine timing of control strategies. In fact, there are recent studies of economic analyses for novel control strategies where economic thresholds are applied in field crops where it is possible to relate number of insects with a damage estimate (Crowder et al. 2006, Antwi et al. 2007, Beres et al. 2007). Indianmeal moth infestation of finished stored products presents a unique challenge in that the products typically are of high-value products and are stored for variable periods of time in multiple locations; thus, the insect threshold is essentially zero. The Indianmeal moth is a cosmopolitan pest known to infest a great number of commodities, including many different

grains, dried fruit and nuts (Mohandass et al. 2006a). Damage caused by Indianmeal moth can range from direct feeding to product contamination, to package holes and ruptures, and to the creation of favorable conditions for mold and bacterial growth. Also, losses from these moths can occur anywhere within the process, from manufacturer to the home of the consumer (Mowery et al. 2004). And, several studies have shown that Indianmeal moth infests facilities ranging from feed mills (Larson et al. 2008) to flour mills and pet food storage facilities (Ryne et al. 2007).

There are many recent examples of the use of pheromone traps, sticky traps, and pitfall traps to monitor insect pests in food storage facilities (Arbogast et al. 2000, Nansen et al. 2004). However, because of the various biological and environmental factors that can affect trap catch, it is often difficult to relate the numbers of insects caught in traps to the actual populations (Arbogast et al. 2002; 2005). Using an economic approach to estimate the need for insecticide applications in food storage sites may be a useful addition to integrated pest management programs. Using target mortality models to analyze economic risks and benefits may enable pest managers to optimize multiple controls and improve their pest management programs.

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Figures and Tables

Table 5-1 Summary by Frequency of Insecticide Application

Number of Treatments in 180 days	0	1	4	6	8	11
Total Cost per	10,000 ft ³ *	\$	\$	\$	\$	\$
No treatment	0	0	0	0	0	0
Methoprene- Surface	0	1.56	6.24	9.36	12.48	17.16
Methoprene- Aerosol	0	0.71	2.84	4.27	5.69	7.82
Methoprene plus Esfenvalerate	0	1.17	4.68	7.014	9.35	12.86
Methoprene plus 1% pyrethrin	0	3.14	12.56	18.85	25.13	34.55

^{*} or 10,000ft² for surface treatments

Table 5-2 Simulations of Survival of Indianmeal moth on Raisins Before and After Insecticide Treatments at 6 Temperatures

Temperature	Insecticide Treatment						
(°C)		No Treatment	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
21	Methoprene plus Esfenvalerate	e 2.E+02	4.E+01	0.E+00	1.E+00	0.E+00	0.E+00
24	Methoprene plus Esfenvalerate	5.E+03	8.E+02	6.E+00	0.E+00	0.E+00	0.E+00
27	Methoprene plus Esfenvalerate	e 2.E+03	4.E+02	8.E+00	0.E+00	0.E+00	0.E+00
30	Methoprene plus Esfenvalerate	e 2.E+03	4.E+02	4.E+00	0.E+00	0.E+00	0.E+00
32	Methoprene plus Esfenvalerate	7.E+02	1.E+02	4.E+00	0.E+00	0.E+00	0.E+00
35	Methoprene plus Esfenvalerate	4.E+00	1.E+00	0.E+00	0.E+00	0.E+00	0.E+00
		No Treatment	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
21	Methoprene plus 1% Pyrethrin	2.E+02	1.E+01	0.E+00	0.E+00	0.E+00	0.E+00
24	Methoprene plus 1% Pyrethrin	5.E+03	3.E+02	0.E+00	0.E+00	0.E+00	0.E+00
27	Methoprene plus 1% Pyrethrin	2.E+03	1.E+02	0.E+00	0.E+00	0.E+00	0.E+00
30	Methoprene plus 1% Pyrethrin	2.E+03	1.E+02	0.E+00	0.E+00	0.E+00	0.E+00
32	Methoprene plus 1% Pyrethrin	7.E+02	4.E+01	0.E+00	0.E+00	0.E+00	0.E+00
35	Methoprene plus 1% Pyrethrin	4.E+00	0.E+00	0.E+00	0.E+00	0.E+00	0.E+00
		No Treatment	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
21	Methoprene- Surface	2.E+02	1.E+02	1.E+01	3.E+00	1.E+00	0.E+00
24	Methoprene- Surface	5.E+03	2.E+03	2.E+02	8.E+01	1.E+01	2.E+00
27	Methoprene- Surface	2.E+03	1.E+03	1.E+02	3.E+01	8.E+00	1.E+00
30	Methoprene- Surface	2.E+03	1.E+03	1.E+02	2.E+01	6.E+00	1.E+00
32	Methoprene- Surface	7.E+02	3.E+02	5.E+01	8.E+00	2.E+00	0.E+00
35	Methoprene- Surface	4.E+00	2.E+00	0.E+00	0.E+00	0.E+00	0.E+00
		No Treatment	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
21	Methoprene- Aerosol	2.E+02	8.E+01	3.E+00	0.E+00	0.E+00	0.E+00
	Methoprene- Aerosol	5.E+03	1.E+03	5.E+01	1.E+01	1.E+00	0.E+00
	Methoprene- Aerosol	2.E+03	7.E+02	4.E+01	4.E+00	1.E+00	0.E+00
	Methoprene- Aerosol	2.E+03	7.E+02	3.E+01	3.E+00	0.E+00	0.E+00
	Methoprene- Aerosol	7.E+02	2.E+02	2.E+01	1.E+00	0.E+00	0.E+00
35	Methoprene- Aerosol	4.E+00	1.E+00	0.E+00	0.E+00	0.E+00	0.E+00

Table 5-3 Simulations of Survival of Indianmeal moth on Wheat Diet Before and After Insecticide Treatments at 6 Temperatures

Temperature	Insecticide						
(°C)	Treatment	No Treatment On	e time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
	Methoprene plus						
21	Esfenvalerate	8.E+08	1.E+08	6.E+06	2.E+05	2.E+04	4.E+02
	Methoprene plus						
24	Esfenvalerate	7.E+10	2.E+10	2.E+10	1.E+07	2.E+06	1.E+04
	Methoprene plus						
27	Esfenvalerate	4.E+12	3.E+12	4.E+10	4.E+10	1.E+08	9.E+06
	Methoprene plus						
30	Esfenvalerate	2.E+14	5.E+13	1.E+12	1.E+11	7.E+09	9.E+07
22	Methoprene plus	1.5.14	2 F. 12	1.5.16	4 E : 10	1.5.11	5 5 . 0 5
32	Esfenvalerate	1.E+14	3.E+13	1.E+12	4.E+10	1.E+11	7.E+07
25	Methoprene plus Esfenvalerate	4 E+06	1.E+06	4 E+04	2 E+02	1 E+02	6 E+00
35	Estenvalerate	4.E+06			3.E+03	1.E+03	
	36.0	No Treatment On	ie time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
21	Methoprene plus	0.5+00	4 E+07	0.5.05	1 5 + 02	0.5+01	0.5.00
21	1% Pyrethrin	8.E+08	4.E+07	9.E+05	1.E+03	9.E+01	0.E+00
24	Methoprene plus 1% Pyrethrin	7.E+10	9.E+09	4.E+07	7.E+04	4.E+03	1.E+00
24	Methoprene plus	7.L+10	9.E+09	4.L+0/	7.L+04	4.L+03	1.E+00
27	1% Pyrethrin	4.E+12	3.E+12	3.E+09	2.E+10	3.E+05	1.E+04
21	Methoprene plus	1.L · 12	3.13 · 12	3.E · 07	2.1.10	3.E · 03	1.1.
30	1% Pyrethrin	2.E+14	3.E+13	8.E+10	4.E+10	1.E+08	4.E+04
	Methoprene plus	2.2 1.	0.2 10	0.2 10	2 10	1.2 00	2 0.
32	1% Pyrethrin	1.E+14	1.E+13	5.E+11	9.E+08	5.E+10	1.E+04
	Methoprene plus						
35	1% Pyrethrin	4.E+06	9.E+05	6.E+03	7.E+01	2.E+02	0.E+00
		No Treatment On	e time	6 Weeks	4 Weeks	3 Weeks	2 Weeks
	Methoprene-						
21	Surface	8.E+08	3.E+08	6.E+07	1.E+07	4.E+06	7.E+05
	Methoprene-						
24	Surface	7.E+10	4.E+10	5.E+09	1.E+09	4.E+08	4.E+07
	Methoprene-						
27	Surface	4.E+12	3.E+12	4.E+11	3.E+11	3.E+10	5.E+09

	Methoprene-						
30	Surface	2.E+14	9.E+13	1.E+13	3.E+12	1.E+12	1.E+11
	Methoprene-						
32	Surface	1.E+14	5.E+13	2.E+12	2.E+12	1.E+12	1.E+11
	Methoprene-						
35	Surface	4.E+06	2.E+06	4.E+05	1.E+05	5.E+04	5.E+03
		No Treatment One	e time 6 W	veeks 4 V	Weeks 3 V	Veeks 2 W	eeks
	Methoprene-						
21	Aerosol	8.E+08	2.E+08	2.E+07	2.E+06	4.E+05	3.E+04
	Methoprene-						
24	Aerosol	7.E+10	3.E+10	2.E+09	2.E+08	4.E+07	4.E + 07
	Methoprene-						
27	Aerosol	4.E+12	3.E+12	1.E+11	1.E+11	3.E+09	4.E + 08
	Methoprene-						
30	Aerosol	2.E+14	7.E+13	5.E+12	7.E+11	1.E+11	7.E+09
	Methoprene-						
32	Aerosol	1.E+14	4.E+13	4.E+12	5.E+11	4.E+11	6.E+09
	Methoprene-						
35	Aerosol	4.E+06	2.E + 06	2.E+05	2.E+04	9.E+03	3.E + 02

Table 5-4 Empirical model with minimization of variable costs for treatments with methoprene plus 1% synergized pyrethrin.

Constraint- Temperature- Diet		Timing of Application					Deviations			Inequality Signs	Mortality Goal
	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks	d1 ⁻	d12 ⁻			
T1 - Wheat Diet	0.00	0.945914	0.998809	0.999999	1.0000	1.0000	1		0.99	≥	0.99
T2 - Wheat Diet	0.00	0.874124	0.999420	0.999999	1.0000	1.0000			0.99	\geq	0.99
T3 - Wheat Diet	0.00	0.360308	0.999156	0.995312	1.0000	1.0000			0.99	\geq	0.99
T4 - Wheat Diet	0.00	0.804419	0.999541	0.999782	1.0000	1.0000			0.99	\geq	0.99
T5 - Wheat Diet	0.00	0.905590	0.996258	0.999993	0.9996	1.0000			0.99	\geq	0.99
T6 - Wheat Diet	0.00	0.797588	0.998629	0.999984	1.0000	1.0000			0.99	\geq	0.99
T1 - Raisins	0.00	0.946939	1	1	1	1			0.99	\geq	0.99
T2 - Raisins	0.00	0.945247	1	1	1	1			0.99	\geq	0.99
T3 - Raisins	0.00	0.944869	1	1	1	1			0.99	\geq	0.99
T4 - Raisins	0.00	0.945514	1	1	1	1			0.99	\geq	0.99
T5 - Raisins	0.00	0.944979	1	1	1	1			0.99	\geq	0.99
T6 - Raisins	0.00	1	1	1	1	1		1	0.99	\geq	0.99
Sum of Deviations							1	1			
Sum Product									1.0		
											\$3.14
Minimum Variable Cost											1

Table 5-5 Empirical model solutions and optimization for treatments methoprene plus 1% synergized pyrethrin.

Model Solution			Timing of Application							
Cost (\$) unit area	Overall Risk Level	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks			
17.68	0	0.01	0.00	0.60	0.13	0.13	0.13			
5.98	1	0.00	0.70	0.30	0.00	0.00	0.00			
2.98	2	0.05	0.95	0.00	0.00	0.00	0.00			
2.68	3	0.15	0.85	0.00	0.00	0.00	0.00			

Table 5-6 Empirical model with minimization of variable costs for treatments with methoprene plus esfenvalerate.

Constraint- Temperature- Diet				Timing of Application			Deviations	Sum Product	Inequality Signs	Mortality Goal
	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks	d1 d12	-		
T1 - Wheat Diet	0.00	0.823227	0.992421	0.999764	1.0000	1.0000	1	0.99	<u> </u>	0.99
T2 - Wheat Diet	0.00	0.760742	0.760742	0.999822	1.0000	1.0000		0.99	≥	0.99
T3 - Wheat Diet	0.00	0.313587	0.990475	0.988878	1.0000	1.0000		0.99	≥	0.99
T4 - Wheat Diet	0.00	0.700078	0.993749	0.999279	1.0000	1.0000		0.99	≥	0.99
T5 - Wheat Diet	0.00	0.788132	0.989063	0.999659	0.9990	1.0000		0.99	≥	0.99
T6 - Wheat Diet	0.00	0.694141	0.990192	0.999374	0.9997	1.0000		0.99	≥	0.99
T1 - Raisins	0.00	0.82449	1	0.995918	1	1		0.99	≥	0.99
T2 - Raisins	0.00	0.823453	0.998707	1	1	1		0.99	≥	0.99
T3 - Raisins	0.00	0.823155	0.996607	1	1	1		0.99	≥	0.99
T4 - Raisins	0.00	0.823391	0.998121	1	1	1		0.99	≥	0.99
T5 - Raisins	0.00	0.822558	0.994498	1	1	1		0.99	≥	0.99
T6 - Raisins	0.00	0.75	1	1	1	1	1	0.99	≥	0.99
Sum of Deviations							1 1			
Sum Product								1.00		
Minimum Variable Cost										\$1.169

Table 5-7 Empirical model solutions and optimization of frequency of treatments for treatments with methoprene plus esfenvalerate.

Model Solution			Timing of Application								
Cost (\$) unit area	Overall Risk Level	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks				
9.24	0	0.01	0.00	0.02	0.39	0.31	0.27				
3.63	1	0.00	0.30	0.70	0.00	0.00	0.00				
2.36	2	0.00	0.66	0.34	0.00	0.00	0.00				
1.16	3	0.01	0.99	0.00	0.00	0.00	0.00				

Table 5-8 Empirical model with minimization of variable costs for treatments with aerosol methoprene alone.

Constraint- Temperature- Diet				Timing of Application	ı		Deviations	Sum Product	Inequality Signs	Mortality Goal
	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks	d1 d12			
T1 - Wheat Diet	0.00	0.690031	0.973377	0.996975	0.9994	1.0000	1	0.99	<u> </u>	0.99
T2 - Wheat Diet	0.00	0.637666	0.976347	0.997395	0.9995	0.9995		0.99		0.99
T3 - Wheat Diet	0.00	0.262839	0.962855	0.972650	0.9992	0.9999		0.99		0.99
T4 - Wheat Diet	0.00	0.586806	0.972361	0.996084	0.9992	1.0000		0.99	_	0.99
T5 - Wheat Diet	0.00	0.660623	0.968851	0.996492	0.9968	1.0000		0.99		0.99
T6 - Wheat Diet	0.00	0.581840	0.964875	0.994690	0.9979	0.9999		0.99		0.99
T1 - Raisins	0.00	0.689796	0.987755	1	1	1		0.99	\geq	0.99
T2 - Raisins	0.00	0.689804	0.989869	0.996982	0.999784	0.999784		0.99	\geq	0.99
T3 - Raisins	0.00	0.689567	0.982188	0.998304	0.999576	0.999576		0.99	\geq	0.99
T4 - Raisins	0.00	0.689995	0.986848	0.998591	1	1		0.99	\geq	0.99
T5 - Raisins	0.00	0.690509	0.976616	0.998624	1	1		0.99	\geq	0.99
T6 - Raisins	0.00	0.75	1	1	1	1	1	0.99	\geq	0.99
Sum of Deviations							1			
Sum Product								1.00		
Minimum Variable Cost										\$0.711

Table 5-9 Empirical model solutions and optimization of frequency of treatments for treatments with methoprene aerosol.

Model Solution			Timing of Application									
Cost (\$) unit area	Overall Risk Level	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks					
6.81	0	0.00	0.03	0.00	0.16	0.10	0.70					
2.40	1	0.00	0.21	0.79	0.00	0.00	0.00					
1.88	2	0.00	0.45	0.55	0.00	0.00	0.00					
1.36	3	0.00	0.69	0.31	0.00	0.00	0.00					

Table 5-10 Empirical model with minimization of variable costs for treatments with methoprene as a surface treatment.

Constraint- Temperatur Diet	·e-			Timing of Application			Deviations	Sum Product	Inequality Signs	Mortality Goal
	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks	d1 ⁻ d12 ⁻			
T1 - Wheat Diet	0.00	0.549946	0.926220	0.982214	0.9948	0.9991	1	0.99	<u> </u>	0.99
T2 - Wheat Diet	0.00	0.508218	0.928429	0.983511	0.9947	0.9994		0.99	_ ≥	0.99
T3 - Wheat Diet	0.00	0.212273	0.901215	0.934323	0.9928	0.9987		0.99	_ ≥	0.99
T4 - Wheat Diet	0.00	0.467671	0.919168	0.981419	0.9929	0.9992		0.99	≥	0.99
T5 - Wheat Diet	0.00	0.598091	0.983633	0.983633	0.9887	0.9991		0.99	<u>></u>	0.99
T6 - Wheat Diet	0.00	0.463724	0.907381	0.974776	0.9886	0.9989		0.99	\geq	0.99
T1 - Raisins	0.00	0.55102	0.95102	0.987755	0.995918	1		0.99	\geq	0.99
T2 - Raisins	0.00	0.549903	0.956672	0.981893	0.997198	0.999569		0.99	\geq	0.99
T3 - Raisins	0.00	0.550042	0.939355	0.98855	0.996607	0.999576		0.99	\geq	0.99
T4 - Raisins	0.00	0.550023	0.949272	0.989197	0.997182	0.99953		0.99	\geq	0.99
T5 - Raisins	0.00	0.550206	0.927098	0.988996	0.997249	1		0.99	\geq	0.99
T6 - Raisins	0.00	0.549946	0.926220	0.982214	0.9948	0.9991	1	0.99	\geq	0.99
Sum of Deviations							1			
Sum Product								1.00		
Minimum Variable Cost										\$1.56

Table 5-11 Empirical model solutions and optimization of frequency of treatments for treatments with surface treatments of methoprene

Model Solution			Timing of Application								
Cost (\$) unit area	Overall Risk Level	None	One time	6 Weeks	4 Weeks	3 Weeks	2 Weeks				
14.91	0	0.00	0.01	0.00	0.00	0.46	0.54				
5.87	1	0.00	0.08	0.92	0.00	0.00	0.00				
4.98	2	0.00	0.27	0.73	0.00	0.00	0.00				
4.09	3	0.00	0.46	0.54	0.00	0.00	0.00				

Summary and Conclusions

Plodia interpunctella Hübner is a major economic pest of a variety of commodities and finished stored products. Pest status is due in part to the fact that it can exploit a wide range of food sources and has a high reproductive capacity. Indianmeal moths can be problematic at all stages of production, storage, and distribution of food products for human consumption and, therefore, cause several types of economic damage. Infestations in manufacturing facilities can damage equipment, primarily through the webbing associated with larval development, as well as cause direct losses in raw and finished products resulting from feeding. Movement of infestations (undetected or unmanaged) from storage to retail outlets may result in consumer complaints and returned products. Indianmeal moth management requires efficient and costeffective management options, while ensuring that these control options are safe to use in facilities where food is produced or stored for human consumption. The insect growth regulator, methoprene, is labeled for a variety of uses, including direct application to stored grains, liquid surface treatments inside and outside a facility, and aerosol applications to the interior of the facility. This insecticide will control many stored product insect pest species, alone, and in combination with conventional insecticides. Insect growth regulators are considered to be "reduced risk" in comparison to neurotoxic insecticides; they are specifically targeted towards insects and have a low risk with respect to non-target organisms, including humans.

In Chapter 1, I examined how temperature and surface substrates affect the efficacy of methoprene applied to control the egg and fifth instars of the Indianmeal moth. Generally, temperature does not affect the efficacy of methoprene applied at the label rate as a surface treatment, but overall development of viable adults from exposed eggs is high in relation to untreated controls. Therefore, methoprene by itself may not be a reliable control strategy with eggs of the Indianmeal moth as the target life stage. Fifth instars appear to be a more susceptible life stage compared to the eggs. When fifth instars are exposed to the label rate of methoprene for at least two hours, survival of the adult stage is reduced compared to untreated controls. Survival is further reduced at exposures of more than two hours, but additional data are needed to assess these situations.

Methoprene applied as an aerosol alone and in combination with the pyrethroid, esfenvalerate, gave a high degree of control of eggs and fifth instars of the Indianmeal moth. These results add to other recent publications demonstrating the efficacy of aerosol treatments for both conventional insecticides and insect growth regulators, and results are discussed in Chapter 2. The other significant result of these studies is that there was no difference in mortality of the target life stages when exposed in the open compared to underneath a shelf, which indicates good coverage and penetration of the aerosol fog. This result is applicable to field conditions, as food manufacturing and storage facilities often have products stacked on pallets which will provide refugial sites for insects underneath the pallets. Results show potential spread and dispersion of aerosols in these obstructed sites.

Installation of a facility-wide aerosol application system, and costs of insecticidal treatments, may be less expensive overall than the costs of frequent fumigation with methyl bromide or any other fumigant. Confirming the effects of aerosol insecticides applied in field systems on Indianmeal moth survival is vital in determination of cost effectiveness of these chemicals. Chapter 3 describes the results of a large-scale study with methoprene delivered as a aerosol, alone and in combination with synergized pyrethrins, in which target life stages were directly exposed or exposed to different treated diets and treated packaging materials. Adult emergence of Indianmeal moth was differentially reduced depending on the diet and packaging product exposed to insecticides. In each of the field trials aerosol particles were effectively distributed underneath stacked product and machinery in each of the field trials. Across all treatment scenarios with the combination of methoprene with synergized pyrethrin, mortality of exposed eggs ranged from 70 to 85%. These results indicate significant effects on the population dynamics of insects infesting these products. Additionally, the most expensive treatment option is not always the lowest risk; in this case, 3% pyrethrin plus methoprene treatment did not reduce survival to the adult stage as much as the other treatments. These results represent the efficacy of these chemicals for eggs of the Indianmeal moth only. Consideration of other pest species and particular needs of each facility must be carefully assessed before insecticide application.

In Chapter 4, model simulations determined how Indianmeal moth populations respond to various temperatures and insecticide treatments on two different diets. Insecticide interventions resulted in a different response on the wheat diet compared to raisins because mean survivorship is much higher on the wheat diet than on raisins. Differences in simulation at different

temperatures would also be expected given that survivorship was modeled in wheat to follow a linear regression, while in raisins the model was developed to fit a quadratic equation. The wheat diet is an example of a "worst case" scenario because it is a product that can support rapid population growth. Populations on the wheat diet were predicted to grow so rapidly that even with an insecticide treatment that produced 95% mortality, population levels rebounded to pretreatment levels within a few days. Aside from the biological parameters (temperature-dependent development time, survivorship and fecundity) placed on the Indianmeal moth populations and the occurrence of insecticide treatment events, the model does not include limits based on the amount or availability of a food source. Despite the simulation of these "worst-case" conditions, populations of Indianmeal moth can grow rapidly given their high reproductive capacity, so management of the Indianmeal moth in any area containing products that contained wheat diet would be crucial to avoid devastating product loss. Overall feasibility in terms of chemical cost and risk related to mortality are presented in each chapter (1-4) in the form of a partial budget analysis.

Treatments in the overall economic model are based on a mix of two diets (wheat and raisins), on which Indianmeal moth survives very well and very poorly. These types of simulations are realistic given that warehouse environments may have many different food types stored in close proximity to one another. Possibilities for simulations are limited only by available pest population growth models and variable cost information. While my simulations and economic models do not include every possible treatment scenario, I have presented a range for which decisions about insecticide type, application type, and frequency could be made under the available model conditions. It may be possible to compare and optimize for non-chemical pest management options such as those already modeled and analyzed, such as chilled air aeration, ambient air aeration fumigation and heat treatments of facilities. My results are especially useful when comparing management strategies such as those listed with costs of low risk insecticides.

Further research must be done to build more exact population growth and economic models related to Indianmeal moth control. One major area where more information is needed is how adults are affected by methoprene. While literature suggests there are sub-lethal effects associated with methoprene, there have been no studies to date on the Indianmeal moth.

As aerosol insecticide treatments becoming increasingly utilized, more field studies must be conducted to assess the effects of these chemicals on other stored product pests.

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