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INTEGRATED MANAGEMENT STRATEGIES FOR CONTROL OF CHERRY FRUIT FLIES, RHAGOLETIS CINGULATA AND RHAGOLETIS FAUSTA (DIPTERA: TEPHRITIDAE)

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Jessica Lynn Kostarides

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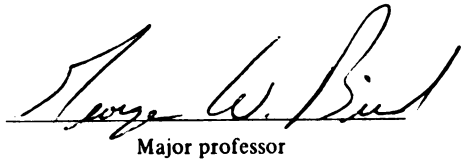

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INTEGRATED MANAGEMENT STRATEGIES FOR CONTROL OF CHERRY FRUIT FLIES, *RHAGOLETIS CINGULATA* AND *RHAGOLETIS FAUSTA* (DIPTERA: TEPHRITIDAE)

By

Jessica Lynn Kostarides

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ABSTRACT

INTEGRATED MANAGEMENT STRATEGIES FOR CONTROL OF CHERRY FRUIT FLIES, *RHAGOLETIS CINGULATA* AND *RHAGOLETIS FAUSTA* (DIPTERA: TEPHRITIDAE)

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The genus *Rhagoletis*, of the dipteran family Tephritidae, includes some 50 described species and is widely distributed over the Holarctic and Neotropical regions. Many of these species are major economic pests of fruit such as cherries. The eastern, *Rhagoletis cingulata* (Loew) and black, *R. fausta* (Osten Saken) cherry fruit flies are native to North America. They are major pests of cultivated sweet *(Prunus avium)* and tart *(P. cerasus)* cherries. In order to ensure maggot-free fruit to meet the stringent zero tolerance levels mandated by Federal and State regulations (FRL-6813-9), growers generally apply two to three applications of broad-spectrum insecticides, primarily organophosphates, for reduction of fly populations. Implementation of the Food Quality Protection Act (FQPA) of 1996 may prevent the future use of organophosphates as well as other conventional insecticides for management of key *Rhagoletis* species. As a result, the focus of my research was to investigate the potential of developing an integrated system for control of cherry fruit flies. This includes: 1) improving current cherry fruit fly monitoring techniques with Rebell™ traps; 2) identifying reduced-risk compounds that can be used in cherry fruit fly management programs; 3) deployment of biodegradable pesticide-treated trapping devices and entomopathogenic nematodes to suppress adult and larval populations respectively. This project has yielded a new set of pest management tools for optimizing control of cherry fruit flies.

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Finally I thank my field assistants, and the staff at the Northwest Horticultural Research Station in Traverse City, MI. It is with their help that made this research possible. This research was supported by grants from Michigan Agricultural Experiment Station and GREEEN Grant and USDA Special Grant.

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INTRODUCTION

Michigan Cherry Industry. Michigan is responsible for a significant portion of national and worldwide cherry production. Michigan currently dominates the tart cherry (*Prunus avium* L.) industry, generating approximately 75 percent of the crop, along with 12 percent of sweet cherries, *P. cerasus* L. In 1999, the gross receipts for both tart and sweet cherries exceeded $50 million (Michigan Agricultural Statistics 1999). There are nearly 40,000 acres of tart cherries in the state with a total of about 55,000 acres in the United States. There are 3.8 million tart cherry trees grown by about 1,000 growers in Michigan. Nationally, there are 5.5 million tart cherry trees and approximately 1,500 cherry growers.

The eastern cherry fruit fly, *Rhagoletis cingulata* (Loew) and the black cherry fruit fly, *R. fausta* (Osten Sacken), are important native pests of Michigan cherries. Both Federal regulations (USDA) and consumers mandate a zero tolerance for maggots in fruit. Rejection of the entire crop from a block of fruit is a standard practice if a single larva is detected in the fruit. Therefore, the economic risks to growers encompass direct loss of yield, loss of export markets, increased control costs, and the expense of constructing and maintaining fruit treatment and eradication facilities. Cherry fruit flies are the most economically important late-season pests of commercially grown cherries in the eastern and midwestern United States (Frick *et al.*, 1954).

Biogeographic Distribution. The genus *Rhagoletis* includes about 50 described species and is widely distributed over the Holartic and Neotropical regions (Bush 1966). The time and place of the origin of the genus *Rhagoletis* is not known. The fact that most

of the species within this genus are adapted to high altitudes or temperate climates supports the theory that the Holartic region is the most probable original center of radiation (Bush 1966). Furthermore, evidence from floristic history of the Holartic region indicates that these species arose sometime within the Oligocene or early Miocene. It is also possible that some of the present sibling species were established during the Pliocene (Bush 1966). This period most likely marks the time when the originally proliferate *Rhagoletis* was broken into three major clades. These clades include one in Asia, another in eastern North America, and a third in Central and South America (Bush 1966). The present distribution of this genus supports this hypothesis.

Two native hosts (*Prunus pennsylvanica* L. and *P. serotina* Ehrh.) of cherry fruit flies generally harbor maggots at severe infestation rates. Both of these wild cherries are widely distributed throughout Michigan's landscape. Cherry fruit flies are well adjusted to these hosts, as is indicated by the heavy infestations of maggots during peak fruiting season.

While both *P. pennsylvanica* and *P. serotina* serve as important native host plants for cherry fruit flies, the host preferences displayed by these insects is unique. Both species of *Rhagoletis* attack the two introduced cultivated cherries (*P. avium* and *P. cerasus*) equally, but their relationship to native wild cherries is more complex. *Rhagoletis fausta* is confined to *P. pennsylvanica*, while *R. cingulata* prefers *P. serotina*. *Rhagoletis fausta*, the species that regularly appears first in cultivated cherry orchards, is associated with the early maturing *P. pennsylvanica* in nature; whereas, *R. cingulata* prefers the later maturing *P. serotina*.

Courtship Behavior and Life Cycle. Cherry fruit flies exhibit a wide array of behaviors throughout their adult life stage. These include dispersal, feeding, and oviposition behaviors, especially in courtship and mating. Males secrete sex-attractant chemicals, either by inflating the lateral abdominal membranes or by extruding an anal pouch, to attract female flies. Wing fanning disperses pheromones, which also precedes sounds of possible significance in courtship. Once on an appropriate host fruit (mating site) males defend their territory with various aggressive displays including head-on collision, "boxing" (fight involving prothoracic legs) and wing jerking, while waiting for females (AliNiazee 1974, Messina and Subler 1995). The female usually visits several fruit prior to oviposition. It spends about 10 days feeding in the vicinity of host fruit and foliage before laying eggs. Oviposition behavior appears to be more uniform than epigamic behavior and consists of the following stages: a) movement towards and arrival at the oviposition site; b) testing the site (for previous oviposition); c) inserting the needle-shaped ovipositor into the fruit; d) and depositing a single egg just below the surface of the fruit. Immediately following egg deposition, the female walks around the fruit dragging her extended ovipositor on the fruit surface, marking it with her pheromone to deter other females from ovipositing in the same fruit, thus preventing competition with the single larva (Prokopy 1976). Each female is capable of depositing 300 to 400 eggs during the three to four weeks she is active. The eggs hatch in about seven days. Legless larvae start to feed and tunnel around to the pit of the fruit in approximately two weeks. There are three to four larval instars, lasting a total of 10 – 21 days. The last instar emerges from the fruit, falls to the orchard floor, and burrows to a depth of 2-5 inches beneath the soil surface.

The majority of temperate *Rhagoletis* spp. are univoltine (Fletcher 1989). Puparial diapause is followed by emergence of adults from the soil beneath host plants that fruited the previous year. Factors that influence emergence of adults include soil moisture, temperature, location, and soil type. Emergence coincides closely with the appearance of host fruit suitable for adult egg deposition. Peak emergence in southwestern Michigan occurs in early to mid June for the black cherry fruit fly and early to mid July in northwestern Michigan for the eastern cherry fruit fly (Liburd *et al.*, 2001).

The major natural enemies of a sibling *Rhagoletis* species, *R. mendax* Curran, are two braconid wasp parasitoids, *Opium ferruginues* Gahan and *O. melleus* Gahan. They emerge from the pupal cases about 25 to 30 days after adult fly emergence (Drummond and Collins 1997b). The parasitoids locate maggots, and overwinter in the blueberry maggot puparium.

Predation is also a key factor in regulation of *Rhagoletis* spp. populations (Boller and Prokopy 1976). Ants, staphylinid beetles, carabids, cecidomyiids, and crickets prey on fruit fly species within the family Tephritidae. Fungi, bacteria, and viruses infect various tephritid species. To date, however, none have been isolated from natural populations of cherry fruit fly (Sivinski 1996). This indicates, therefore, a necessity to investigate alternative biological control methods for suppression of *Rhagoletis* spp.

Nematode Classification and Biology. Nematode parasites of insects have been known since the 17th century (Nickle 1984), but it was only in the 1930s that serious consideration was given to using a nematode to control insects. Nematodes are vermiform, non-metametrically segmented invertebrates with bilateral symmetry. They have a digestive system with a three-angled esophageal lumen. Nematodes have separate sexes. Males have spicules and tubular gonands joining the digestive system to form a cloaca, and females with both a gonopore and a posterior opening to the digestive system. Numerically, they are the most abundant multicellular organisms on the planet (Bird 2002, personal communication). Some species function as bacterivores, whereas others feed as fungivores, algavores, herbivores or omnivores.

Species in the families *Steinernematidae* and *Heterorhabditidae* (Rhabditida: Nematoda) parasitize insects. These are the most economically important entomopathogenic nematodes. As beneficial organisms they are lethal to many important soil insect pests and yet are safe for non-target arthropods.

Infective juveniles of *Steinernema* spp. are 0.5 mm to 1.5 mm in length depending on the species. They have stomal and anus plugs and cannot feed until a host is available. Usually the infective juveniles found in soil, are activated by insect movement and then follow a gradient of carbon dioxide to find the insect (Gaugler and Kaya 1990). Entomopathogenic nematodes enter through natural body openings, the mouth, anus or respiratory inlets (spiracles) and then penetrate into the blood cavity from the gut or breeding tubes (Poinar 1990). *Heterorhabditis* spp. can also penetrate through the insect's interskeletal membranes by scratching away at these with a stomatal tooth (Bedding and Molyneux 1982).

Once in the insect's blood, infective juveniles release a highly specialized symbiotic bacterium found only in entomopathogenic nematodes. These are *Xenorhabdus* spp. in *Steinernema* spp., and *Photorhabdus* spp. in *Heterorhabditis* spp. Bacteria multiply, produce toxins, and kill the insect within approximately one day. As decomposers, bacteria degrade the insect, and replicate, forming food for the nematode. They also produce a range of antibiotics (Akhurst 1982) and anti-feedants that preserve the dead insect while the nematodes feed and breed within it.

Because entomopathogenic nematodes require continual oxygenation, it is impractical to supply them as biological control agents in a water suspension that would have to be kept continually aerated and would even then only last for a few days. Temporary refrigerated storage can be achieved by adding a cream of nematodes to crumbed foam in plastic bags (Bedding 1984) but the consumer has to extract these from the foam. Further research, however, has led to more suitable means of formulating entomopathogenic nematodes (Bedding 1986, Bedding and Butler 1994, Wang and Bedding 1998). The latest formulation is comprised of about 50% micro-cellulose and 50% nematodes that can be readily mixed in spray tanks, sprayed without blocking nozzles and can survive several months at room temperature. This long shelf life has been achieved by manipulating the nematodes' physiology. Water is removed from between the nematodes over a filter under vacuum and then more water (about 50 %) is removed from within the nematodes by mixing them with just the right amount of dry absorbent. This causes the nematodes to increase their carbohydrate reserve about 10 % prior to entering into hibernation. While in hibernation, they may use only one hundredth of the oxygen that non-hibernating nematodes do and in theory at least can last 100 times

as long. One problem with this is fungal contamination. It has been very difficult to find a suitable preservative that does not harm the nematodes while providing fungal degradation at bay for several months. Another problem is that the nematodes must be kept at precise water content for maximum shelf life. It is also difficult to provide them with oxygen while preventing water loss.

Michigan Monitoring and Management Programs. For the past three decades, fruit fly management programs in Michigan have primarily relied on the use of visual and olfactory traps to accurately time insecticide applications. Strategies adapted by most commercial growers involve hanging baited Pherocon AM yellow sticky boards (Great Lakes IPM, Vestaburg, MI) in orchards. After detection of a single fly, ground or aerial applications of insecticides are made (Liburd *et al.* 2001). A major problem with these traps is that after two weeks of deployment, the sticky boards become inundated with a wide variety of non-target insects, reducing their effectiveness (Liburd *et al.*, 1999). In addition, the odor of the decomposing insects within the Tangle–Trap® interferes with the insect attractant (ammonium compounds) used with these traps, thus reducing their effectiveness. These circumstances dictate that traps be cleaned and replaced on relatively short intervals. Trap preparation, replacement, and maintenance are a time-consuming and labor-intensive operation (Prokopy *et al.,* 1990). Substantial improvements in trapping techniques are needed if traps are to be used in the future for effective monitoring and control programs in commercial cherry production.

Federal laws mandate a zero tolerance for maggot-infested fruit in cherries at harvest. In order to meet these stringent tolerance standards, fruit fly management

programs in Michigan have relied heavily on prophylactic sprays of broad-spectrum organophosphate (OP) compounds. Registered insecticides for fruit fly control include malathion, carbaryl, phosmet, pyrethrin, and azinphos-methyl. Currently, commercial growers apply two to four scheduled sprays of organophosphate insecticides against the cherry fruit flies, irrespective of the presence of adults (Liburd *et al.,* 1998). Organophosphate insecticides are generally effective. They can, however, negatively affect beneficial insects and other non-target organisms, particularly invertebrates in the immediate and surrounding habitats. The results can have a significant negative impact on the full potential of biological control. The future use of organophosphates for management of fruit flies is likely to be restricted because of the 1996 Food Quality Protection Act (FQPA). This law requires the Environmental Protection Agency (EPA) to re-evaluate health risks from pesticides in the U.S.

Insecticide History. The development of concepts for control of fruit-feeding *Rhagoletis* spp. reflects the history of insect control in general. Prior to the 1940's, the insecticidal value of a number of inorganic chemicals (i.e. arsenic) and organic chemicals of botanical origin (i.e. pyrethrum and nicotine) was known, and they were extensively used.

One of the first synthetic organic insecticides was DDT, a halogenated hydrocarbon. In 1877, Zeider synthesized DDT. This chemical, however, was not developed as an insecticide until it was rediscovered by Müller in 1936. Dr. Paul Müller of the Geigy Company of Switzerland developed DDT in 1939. Organochlorine molecules tend to be relatively stable because of the placement of the chlorine ions in the

molecule. Most of these chemicals have been banned because of their persistence in the environment and toxicity to non-target organisms. Soon after DDT was released into the market in the early 1940's, primarily to control lice and fleas that were vectoring disease organisms to humans in the war-torn Europe, the organophosphorous insecticides were developed.

Dr. Schrader of I. G. Farben in Germany (1934) discovered that certain organophosphorous compounds as insecticides. They were initially developed as nerve gasses for chemical warfare. Their toxic mode of action kills insects and vertebrates by binding with acetyl cholinesterase in the synaptic junctions of the nervous system. The resulting effect is a continuous electrical "firing" of chemical signals along the nerve, and repeated muscle contraction and death by exhaustion. Organophosphates (i.e. azinophos-methyl) are widely used for fruit fly control in current cherry production systems.

In 1962, Rachel Carson's book *Silent Spring*, shook public confidence in pesticides. She painted a grim picture of environmental consequences of careless pesticide use. Although the quality of her reporting has been severely criticized, Carson, more than anyone before, increased awareness to the risks of pesticides. This increased public awareness has led to a redirection of public policy as well as research toward more reduced-risk pesticides and cropping methods that reduce the reliance on pesticides.

Since many of the halogenated hydrocarbons, i.e. organophosphates, are being re-evaluated, restricted or eliminated, it is of utmost concern for researchers to explore alternative control methods. Therefore, new classes of chemicals with promising insecticidal properties are being actively evaluated for pest management. Two promising classes presently being studied in lab and field trials include the neonicotinoids (i.e.

imidacloprid) and naturalytes (i.e. spinosyns). Imidacloprid is a systemic and contact insecticide developed by Bayer AG of Germany. It has low toxicity to vertebrates and is seen as a safe alternative to organophosphates and carbamates (CB's). Imidacloprid exhibits a wide range of activity against insects, including members of the order Diptera. Nicotinergic acetylcholine is a neurotransmitter in the synaptic junction of the cholinergic system of insects, and imidacloprid blocks the binding of this neurotransmitter to its postsynaptic receptor. Imidacloprid is very stable in the soil, having a half-life of about 150 days. However, it has rapid ultra-violet (UV) break down and is relatively immobile in the soil and not likely to be a contaminant of ground water.

The spinosyns are a naturally derived group of chemicals produced from the newly discovered actinomycete species, *Saccharopolyspora spinosa*. The discovery and characterization of this soil actinomycete represents a novel opportunity to develop a portfolio of progressive insect management tools. Spinosad was isolated from an organism found in soil samples taken in the Caribbean in 1982 by Dow AgroSciences. To determine its commercial potential, researchers tested the organism and the metabolites it produced during fermentation. It was discovered that the organism produced active metabolites that provided excellent insecticidal activity. Spinosad is a mixture of spinosyn A & D, produced by *S. spinosa* (Kirst *et al.*, 1992). Spinosad affects insects in a range of orders, including Diptera. This biological insecticide demonstrates rapid contact and ingestion activity in insects, which is unusual for a biological product (Larson *et al.*, 1994). It generally controls pest organisms more rapidly than do compounds that depend solely on ingestion. This material degrades rapidly in the

environment, primarily due to photolysis, and has low toxicity to beneficial insects. It is a nerve poison; however, and the actual mode of action is unknown.

History of Pesticide Regulation. The Federal government first regulated pesticides when Congress passed the Insecticide Act of 1910. This law was intended to protect farmers from tainted products. Congress broadened the federal government's control of pesticides by passing the original Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) in 1947. FIFRA required the Department of Agriculture to register all pesticides prior to their introduction in interstate commerce. A 1964 amendment authorized the Secretary of Agriculture to refuse registration to pesticides that were hazardous or ineffective and to remove them from the market. In 1970, Congress transferred the administration of FIFRA to the newly created EPA. This was the initiation of a shift in the focus of federal policy from the control of pesticides for reasonably safe use in agricultural production, to the control of pesticides for reduction of unreasonable risks to man and the environment. This new policy focus was expanded by the passage of the Federal Environmental Pesticide Control Act of 1972 (FEPCA), which amended FIFRA by specifying methods and standards of control, in greater detail. In 1996, Congress unanimously passed a landmark pesticide food safety legislation supported by the Administration and a broad coalition of environmental, public health, agricultural and industry groups. President Clinton promptly signed the bill on August 3, 1996, and the FQPA became law.

The EPA regulates pesticides under two major federal statutes. Under FIFRA, the EPA registers pesticides for use in the United States and prescribes labeling and other

regulatory requirements to prevent unreasonable adverse effects on health or the environment. Under the Federal Food, Drug, and Cosmetic Act (FFDCA), the EPA establishes tolerances (maximum legally permissible levels) for pesticide residues in food. For over two decades, there have been efforts to update and resolve inconsistencies in the two major pesticide statutes, but consensus on necessary reforms remain elusive. The Food Quality Protection Act (FQPA) represents a major breakthrough, amending both major pesticide laws to establish a more consistent and comprehensive, protective regulatory scheme, grounded in sound science.

The FQPA charges the EPA with developing and implementing regulations to enhance protection of the U.S. food supply from pesticide risks also protection of agricultural workers, who apply these compounds. A key provision of the Act calls on the EPA to evaluate pesticide residue risks based on aggregate exposure to all pesticides that share a common toxicological effect on humans. Initial moves by the EPA raised concerns in the agricultural community that FQPA implementation might result in sudden bans on broad classes of pesticides that have been key to U.S. farm productivity. However, the EPA has taken a proactive stance to eliminating label uses. The EPA is allowing some chemistries to remain in use until viable alternatives are available. The viability of the replacement is often debated, and the EPA is aware of the devastating impact that wholesale bans of effective chemistries would have.

The EPA's Office of Pesticide Programs has identified three classes of pesticides as being high risk to human health, and these are receiving the primary scrutiny under the FQPA. They include the organophosphate and carbamate insecticides and the group of broad-spectrum chemicals (known as B-2 chemicals) classified as potential carcinogens.

Almost all of these pesticides are used in current cherry (fruit) Integrated Pest Management (IPM) programs.

Pest management in fruits, vegetables, and both human and animal health programs offers few alternatives to pesticides. There are no immediate replacements for the use of OPs, CBs, or B-2 fungicides for some crops. In addition, the cost of the alternatives, and the difficulty of related operational dynamics and adoption complexities pose a significant threat to the survival of an industry. Under the FQPA, the continued viability of many minor crops will depend on the ability of growers to transition to new pest control tools and knowledge-intensive, site-specific IPM programs.

In the case of perennial fruit crops, such as cherries, growers might be able to adopt a range of OP, CB, and B-2 fungicide mitigation processes including extended pre-harvest application intervals and post-harvest residue removal processes. Other less reliable and more complex alternatives, like pest species-specific pheromones to disrupt insect mating, insect growth regulators, synthetic pyrethroid insecticide substitutes, new fungicides, and pathogen-resistant varieties, may provide some relief. Yet, established plantings cannot be changed quickly without high costs, market dislocation, and new variety acceptance on the part of consumers. Even where some alternatives are available, many producers may not be able to adapt quickly enough to service these sudden changes.

Many existing IPM programs have natural and/or augmentative biological control organisms that have evolved resistance to the OP and/or CB insecticides. Rapid change from an OP-based IPM program to synthetic pyrethroids may provide an alternative for some pests, but will likely result in secondary pest outbreaks as natural enemies are

killed. Each pest-crop and local/regional crop situation will present a different challenge for IPM managers. Clearly, alternatives for one pest-crop situation may not be suitable for another pest, crop, local area, or region.

Where key pesticides are unlikely to be reregistered under the FQPA and where cancellation is imminent, both the USDA and the EPA are working cooperatively, as mandated, to ameliorate any short- or long-term dislocation in local crop production and rural economic stability. This situation will demand a new local-area, alternative IPM program requiring significant additional public sector support for development and implementation of replacement IPM programs. In general, the sudden elimination of key broad-spectrum pesticides that share a similar toxicological effect on humans could cause serious economic hardship to U.S. fruit farmers. Especially where few alternatives exist to the OP, CB, and B-2 fungicides, elimination of one or more of these pesticides groups could cause production of certain crops to shift away from traditional production regions; the results may be a shift to imported foods from countries that have less restrictive agricultural pesticide policies.

Research Objectives. As Federal mandates on the use of organophosphates become increasingly more stringent, the need to develop environmentally sound management programs for cherry fruit flies is of utmost concern. Our research goal was directed towards developing an improved monitoring system for both species of cherry fruit flies and finding alternative strategies for reducing the use of organophosphates in commercial cherry orchard systems. The specific project objectives were 1) To study insect behavior and refine monitoring programs; 2) To identify reduced-risk compounds

that can be used in cherry fruit fly management programs; 3) To investigate the potential of integrating biodegradable pesticide-treated spheres and entomopathogenic nematodes to significantly reduce grower's reliance on organophosphates.

Our preliminary hypothesis was that improving upon current monitoring techniques would enable growers to better predict the primary flight of cherry fruit flies, and infestation levels. This would enable growers to make sound decisions in fruit fly treatment programs. This, accompanied by reduced-risk insecticides and/or biodegradable pesticide-treated trapping devices will ultimately reduce reliance and deposition of harmful and unnecessary spray applications. Furthermore, incorporating natural enemies, such as entomopathogenic nematodes, would suppress larval and pupal cherry fruit fly populations, thereby lowering infestation levels in fields. The net effect of this project will be a new set of pest management strategies for control of cherry fruit flies.

CHAPTER ONE

VISUAL AND OLFACTORY STIMULII AFFECTING THE RESPONSE OF CHERRY FRUIT FLIES (DIPTERA: TEPHRITIDAE).

INTRODUCTION

Passage of the 1996 Food Quality Protection Act (FQPA) and environmental awareness have provided an impetus for producers, as well as other agricultural personnel, to reduce the amount of pesticide used on farms throughout the United States. One of the ways to accomplish this reduction in pesticide usage is to improve monitoring protocols to detect the presence of key pests, so that pesticides are used judiciously when pests are present and when alternative management tools are not available.

The eastern cherry fruit fly, *Rhagoletis cingulata* (Loew), and black cherry fruit fly, *R. fausta* (Osten Sacken), are key late-season pests of cultivated cherries, *Prunus* spp. in the eastern and midwestern United States. Their larvae develop inside the fruit, causing major tissue damage. Entire shipments of cherries can be rejected if one or more maggots are found (Liburd *et al.*, 2001). In order to ensure maggot-free fruit growers, generally apply broad-spectrum prophylactic sprays on a 2-3 week calendar basis (Edson *et al.*, 1998). These insecticides are generally effective, but they could potentially affect non-target organisms, particularly invertebrates in the immediate and surrounding habitats.

Since the 1980s, cherry fruit fly IPM programs in Michigan commercial orchards have relied on visual and olfactory traps for monitoring the presence of adult cherry fruit flies. Until recently, the standard trap used by most growers has been the Pherocon AM yellow board (Great Lakes, IPM) deployed in a vertical orientation (Prokopy 1975; Reissig 1976). However, in a recent paper, Liburd *et al.* (2001) showed that an unbaited three-dimensional Rebell™ trap (Swiss Federal Research Station, Wadswill, Switzerland)

was two times better than Pherocon AM yellow boards in detecting adult cherry fruit flies. Unbaited Rebell™ traps used in their study captured cherry fruit flies 1-2 wk earlier and were more selective for both eastern and black cherry fruit flies.

In previous studies with the related European cherry fruit fly, *Rhagoletis cerasi* (L.), Russ *et al.* (1973) found that the efficiency of a three-dimensional trap was superior to that of the two-dimensional Pherocon AM or rectangular boards. Additional investigations found that daylight fluorescent yellow was also highly attractive to *R. cerasi*.

Rhagoletis cingulata and *R. fausta* respond differently to host stimuli. Howitt (1993) found that *R. cingulata* prefers to forage and oviposit in sweet cherries *P. avium*. Whereas *R. fausta* favors sour cherries (*P. cerasus*). Smith (1984) recorded a preference for fruit compared with leaves for *R. cingulata*, while Prokopy (1976) noted that *R. fausta* spends more time on the leaves compared with the fruit of host plants.

In addition to preferences for host species, other factors are known to affect the behavior of tephritids. Recently, Thornton and Liburd (1999) noted that various habitat-associated factors, including the abundance of wild hosts and rainfall, affected the number of *R. cingulata* found in cherry orchards in northwestern Michigan. Other studies have shown that the state of susceptibility of the fruit affected the ability of *R. mendax* Curran to oviposit in blueberries (Liburd *et al.*, 1998). Moreover, in apples, the ripening phenology and degree of hardness affected the oviposition rate of *R. pomonella* (Messina and Jones 1990). Roitberg *et al.*, (1982) and Averill (1996) found that the presence of oviposition scars decreased *R. pomonella*'s decision to oviposit into fruit.

Our objective was to study the visual and olfactory responses of *R. cingulata* and *R. fausta* using baited and unbaited Rebell™ traps. This goal was to build upon previous research by improving monitoring efficacy and determining the effects of host stimuli on the abundance of cherry fruit flies. The specific objectives were to: 1) to determine whether baiting Rebell™ traps would increase the captures of cherry fruit flies, and 2) Determine the roles of fruit and foliage in attracting black cherry fruit flies to commercial orchards.

MATERIALS AND METHODS

Rebell™ Experiment. Experiments to evaluate baited versus unbaited Rebell™ traps were conducted at the Michigan State University Northwest Horticultural Research Station (NWHRS), in Traverse City, Michigan, and at an abandoned sour cherry, *P. cerasus,* orchard in southwestern Michigan. At the NWHRS, the experiment was conducted from 1 July to 25 August 2000. In southwest MI, the experiment was conducted from 29 May to 2 July 2001. Each experimental site consisted of a 3-hectare block of non-sprayed cherries. The NWHRS site has a dominant residential population of *R. cingulata*, whereas the southwestern site has a high resident population of *R. fausta* (Liburd *et al.*, 2001).

Prior to deploying Rebell™ traps in the orchard, all traps were washed with Histoclear® (Great Lakes IPM, Vestaburg MI) to remove tangle-trap, rinsed twice with distilled water, and allowed to air dry. The traps were then sprayed with a thin layer of insect Tangle-Trap® (aerosol formula, Tanglefoot Company, Grand Rapids, MI). Baited traps had a green polycon dispenser containing 5.0 g of ammonium acetate attached to the top surface of the trap.

The experimental design was a randomized complete block with four replicates. Traps were hung within the center of the tree canopy and spaced 20 m apart within rows, and approximately 30 m between blocks. The foliage immediately surrounding the traps was cleared to prevent any interference between the traps and tree canopies (Drummond *et al.,* 1984). All traps were rotated within blocks on a weekly basis (Fig.1).

Four treatments consisting of the three-dimensional Rebell™ trap were evaluated: 1) baited yellow Rebell™; 2) unbaited yellow Rebell™ used by Liburd *et al.*, (2001); 3) unbaited transparent (made from plexiglas) Rebell™; and 4) baited transparent Rebell™.

Fruit versus Foliage. Experiments to investigate the relationship between host plant characteristics and the abundance of black cherry fruit flies were conducted at the sour cherry, *P. cerasus*, site adjacent to our abandoned cherry fruit fly Rebell™ trap experiment in southwestern, Michigan. A one-hectare block of unsprayed cherries was used for this experiment. The experimental design was a completely randomized design with four replications. Three treatments were evaluated: 1) cherry trees with fruit and foliage, 2) cherry trees with fruit and no foliage, and 3) cherry trees without fruit, but with foliage. Cherry trees without leaves and with fruit were defoliated manually prior to the start of the experiment. Our defoliation process continued on a weekly basis to ensure there was no leaf growth. Trees with leaves and without fruit were de-fruited in a similar fashion to our leaf defoliation process. An unbaited Rebell™ trap was hung in the center of the each test tree canopy to monitor fly population.

Sampling. *R. cingulata* and *R. fausta* caught on traps were counted by sex two times per week, and trapped flies were removed. Female flies caught on traps were classified as mature or sexually immature after thorough examination of the rear abdominal segments.

Statistical Analysis. Data from all experiments were square-root transformed ($x + 0.5$) and then subjected to an analysis of variance (SAS Institute 1989). Least Significant Difference Test (LSD) was used to identify differences in treatment means ($P \leq 0.05$). The untransformed mean values are presented in the Tables and Figures.

RESULTS

Rebell™ Experiment. 2000. In experiments to investigate the visual and olfactory responses of *R. cingulata*, significantly *($F = 77.2$; df = 3, 9; $P < 0.01$)* more cherry fruit flies were caught on the baited Rebell™ traps compared with the other traps tested (Fig. 2). Baited Rebell™ traps captured 1.8, 8.6, and 18.8 times as many *R. cingulata* as the unbaited Rebell™, baited transparent, and un-baited transparent Rebell traps, respectively (Fig. 2). The unbaited Rebell™ trap (standard) caught significantly more *R. cingulata* than the baited or unbaited transparent traps. On an average, unbaited traps captured 4.9 times as many flies than the transparent traps (baited and un-baited) (Fig. 2).

During the 2000 field-season, the first *R. cingulata* was captured on June 30 and peak activity was recorded on July 10 (Fig. 3). After July 10, *R. cingulata* activity declined rapidly and no flies were caught after August 25 (Fig. 3). Yellow Rebell™ traps (baited and unbaited) captured significantly ($F = 42.9$; df = 1,3; $P < 0.01$) more *R. cingulata* females than males (Table 1). Both baited and unbaited yellow Rebell™ traps caught 2.6 and 3.7 times as many females as males, respectively (Table 1). More than 70% of females caught were sexually mature. There were no significant differences for females and males between baited and unbaited transparent Rebell™ traps (Table 1).

2001. The responses of *R. fausta* to baited and un-baited Rebell™ traps differed from those observed for *R. cingulata*. There was no significant difference between baited and unbaited (standard) yellow Rebell™ trap captures (Fig. 4). However, these two traps (baited and unbaited yellow Rebell™) captured significantly ($F = 12.5$; df = 3,9; $P < 0.01$) more flies than transparent traps (Fig. 4). On average, baited and unbaited yellow traps

caught 13.8 times more flies as transparent Rebell™ traps (Fig. 4). There were no significant differences between baited and unbaited transparent traps (Fig. 4).

Rhagoletis fausta began emerging on May 24 when ~ three flies were caught on baited Rebell™ traps (Fig. 5). Trap captures quickly increased and peaked within three weeks on June 14 (Fig. 5). An average of 140 and 160 flies were captured on baited and unbaited Rebell™ traps, respectively (Fig. 5). After peak emergence, fly captures declined slowly and no flies were captured after July 4 (Fig. 5). Both baited and unbaited yellow Rebell™ traps captured significantly ($F = 19.2$; df = 1,3; $P < 0.01$) more females than males *R. fausta* (Table 2). Approximately 75% of the females caught were sexually mature. There was no significant difference in ratio of female to male captures of *R. fausta* with baited and unbaited transparent Rebell traps (Table 2).

Fruit and Foliage. Our results showed that sour cherry trees *P. cerasus* with adequate fruit and leaves were significantly ($F = 12.0$; df = 2,6; $P < 0.01$) more attractive to *R. fausta* than trees that had fruit but no leaves, or trees that had leaves but no fruit (Fig. 6). Unbaited yellow Rebell™ monitoring traps placed within cherry trees with fruit and foliage caught 1.5 and 31.5 times as many flies as traps placed in trees with fruit but no leaves and no fruit but with leaves respectively (Fig. 6). Trap captures were also significantly ($F = 16.2$; df = 2,6; $P < 0.01$) more abundant for *R. fausta* in cherry trees that had fruit but no leaves compared with trees with no fruit but with leaves. Traps placed in sour cherry trees deprived of leaves (with an abundant fruit supply) captured 16.5 times as many flies as traps placed in trees without fruit but with leaves.

DISCUSSION

Response of Cherry Fruit Flies to Rebell™ Traps. Findings indicate that ammonium acetate-baited yellow Rebell™ traps were considerably more effective in detecting the presence of eastern cherry fruit flies compared with other traps evaluated in the study. Possible implications of these observations on baited versus unbaited yellow Rebell™ traps and the two species of cherry fruit flies have interesting trends in relation to host and fly maturity. A major concern regarding ammonium-baited traps is that they attract non-target organisms (Liburd *et al.*, 2000, Drummond *et al.*, 1984), which may be important in regulating cherry fruit fly populations.

Although there was hardly any attraction of *R. cingulata* to baited transparent Rebell™ traps, adding a visual (yellow color) stimulus significantly increased trap captures above the standard unbaited yellow Rebell™, which may suggest that both visual (yellow) and olfactory (bait) stimuli are responsible for the increased attraction of *R. cingulata* to the baited yellow Rebell™ traps. Previous work by Liburd *et al.*, (2001) investigated the potential for using unbaited yellow Rebell™ traps for monitoring *R. cingulata* and *R. fausta* populations.

In our studies, *R. fausta* responses were different from those of *R. cingulata*. The fact that both baited and unbaited yellow Rebell™ traps were considerably more attractive to *R. fausta* than transparent (baited and unbaited) traps suggests that color is more important than odor. Yellow color plus ammonium acetate bait may act synergistically in one species (Eastern) but not synergistically in the other (black). Daylight fluorescent

yellow has previously been shown to be the principal attractant influencing the response of the related European cherry fruit fly, *R. cerasi,* to three-dimensional trapping devices (Russ *et al.*, 1973).

Evidence for behavioral differences between *R. cingulata* and *R. fausta* have been reported earlier by Liburd *et al.*, (2001). In their studies, *R. fausta* trap captures for the unbaited yellow Rebell™ traps were higher than several types of baited and unbaited commercial traps they evaluated. When the same study was repeated with *R. cingulata*, captures from unbaited yellow Rebell™ traps were not significantly different from baited yellow boards and red spheres, demonstrating clear behavioral differences between the species. Our study supports the hypothesis that *R. cingulata* is highly responsive to ammonium baited yellow Rebell™ traps, whereas with *R. fausta* there is no statistical difference between ammonium-baited Rebell™ traps and the standard Rebell™ trap.

Approximately 2.8 times as many females compared with males were caught throughout the season. I presume that more sexually mature females were caught on yellow Rebell™ traps because they are moving into commercial cherry orchards where abundant resources are available for oviposition and larval development. Sexually mature females are probably moving into commercial orchards from adjacent woodlands where wild hosts may be limited.

It is important to note the difference in peak trapped populations for the eastern cherry fruit fly (Fig. 3). Populations peaked approximately 7 July for baited Rebell™ traps, and then dropped off through the end of the season. Interestingly, trapped populations were rapidly declining for the baited Rebell™, while increasing for yellow unbaited Rebell™ traps. This raises the question of the bait potentially being a feeding

deterrent at this point in time of *R. cingulata* development. Laboratory assays to study *R. cingulata* behavior is recommended to investigate this observation.

In this study, there was evidence that females may be better at detecting color than males. This was clearly demonstrated when no differences between females and males captures for the transparent Rebell™ traps were detected while significantly more females than males were captured on yellow Rebell™ traps.

Fruit and foliage. In studies investigating the effects of presence or absence of fruit and leaves, we found that a greater number of flies were foraging in trees that had an abundant supply of fruit and foliage. Host fruit provide adequate resources for egg deposition and larval development for *Rhagoletis* species, and leaves provide sites for feeding and shelter (Prokopy 1976). When both fruit and leaves are in abundant supply, they provide optimum resources for *R. fausta* diurnal activities. This was evident in our studies since host trees without fruit and with an abundant supply of leaves had considerably fewer flies on traps. Previous studies have indicated that *R. fausta* visit leaves more frequently than fruit (Prokopy *et al.,* 1976). However, our work indicates that fruit is more important than leaves to sustain a high population of *R. fausta* flies. Fruit maturity, fly maturity, temperature and rainfall can also affect the number of flies foraging in cherry orchards (Thornton and Liburd 1999).

These results have important implications with respect to monitoring cherry fruit fly populations. When researchers, growers and agricultural extension personnel are developing monitoring protocols for *R. cingulata* in cultivated cherries, they should consider using an ammonium bait to improve early detection of *R. cingulata*. In

Michigan, these baited yellow Rebell™ traps should be deployed in mid-June prior to *R. cingulata* emergence. However, since there is no significant response difference of *R. fausta* to baited yellow Rebell™ traps and the standard Rebell traps, growers can implement the standard unbaited yellow Rebell™ traps for monitoring *R. fausta* populations. These unbaited yellow Rebell™ traps should be deployed in Michigan cherry orchards during mid-May before *R. fausta* emerges.

Table 1. Comparison of total male and female *Rhagoletis cingulata* captures on four types of traps, Traverse City, MI (2000).

	Mean no. of *Rhagoletis cingulata* captured ± SEM	
Treatments	**Females**	**Males**
Baited Rebell™	182.5 ± 19.7a[1]	70.3 ± 3.9b
Rebell™	115.0 ± 19.7a	31.3 ± 6.2b
Baited Transparent	20.5 ± 3.8a	12.8 ± 0.8a
Transparent	13.5 ± 7.4a	7.0 ± 3.1a

[1]Row means followed by the same letter are not significantly different ($P = 0.05$, LSD test).

[2] Statistical analyses were done on the square-root transformed data.

Table 2. Comparison of total male and female *Rhagoletis fausta* captures on four types of traps, Paw Paw, MI (2001).

	Mean no. of *Rhagoletis fausta* captured ± SEM	
Treatments	Females	Males
Baited Rebell™	503.0 ± 55.12a[1]	90.5 ± 16.1 b
Rebell™	587.3 ± 18.6 a	101.0 ± 21.6b
Baited Plexiglas	41.3 ± 2.7 a	6.1 ± 2.2 a
Plexiglas	13.8 ± 2.9 a	4.3 ± 2.2 a

[1]Row means followed by the same letter are not significantly different ($P = 0.05$, LSD test).

[2] Statistical analyses were done on the square-root transformed data.

Trap Type

BR = Baited Rebell™
R = Rebell™
BT = Baited Transparent
T =Transparent

Rep 1 Blk 1	Rep 2 Blk 2	Rep 3 Blk 3	Rep 4 Blk 4
BR	R	T	BT
BT	BR	R	T
R	T	BT	BR
T	BT	BR	R

Fig. 1. Completely randomized block design for modified Rebell™ experiment; treatments rotated within block on a weekly basis.

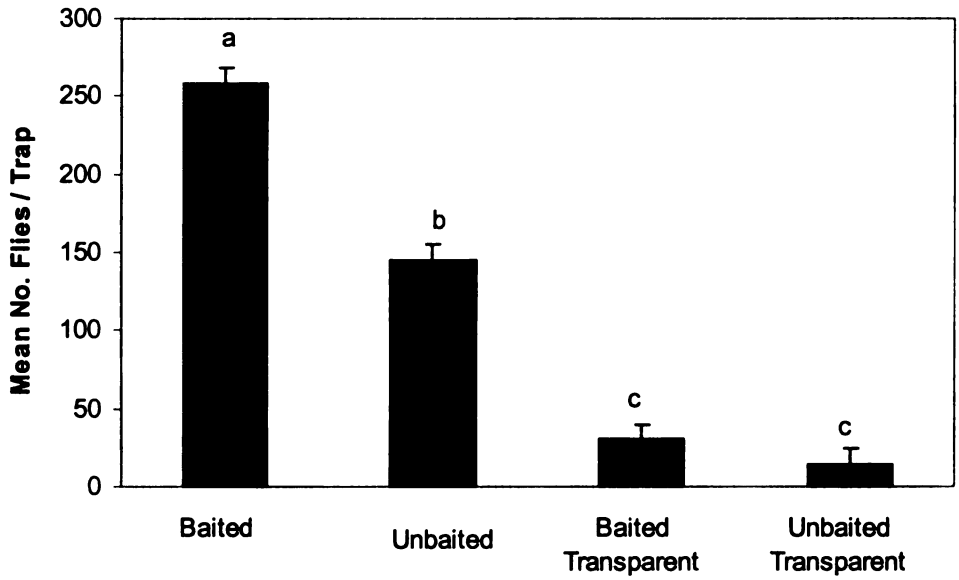

Fig. 2. Total *Rhagoletis cingulata* captures 1 July – 25 Aug. on four types of modified Rebell™ traps, Traverse City, Michigan (2000).

($F = 77.2$; df = 3, 9; $P < 0.01$)

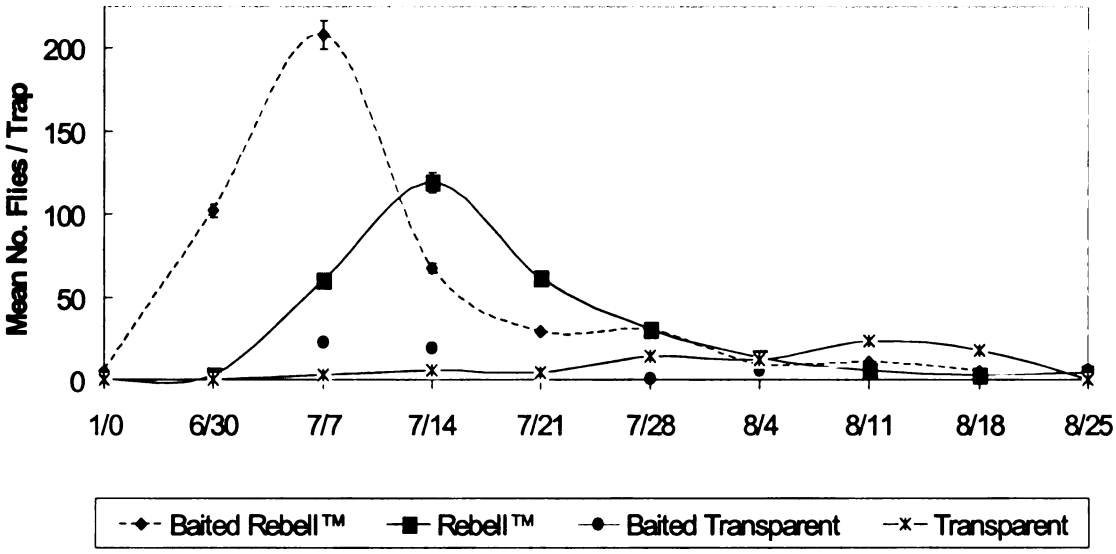

Fig. 3. Comparative population dynamics of *Rhagoletis cingulata* as measured by four trapping systems in a Traverse City, MI cherry orchard in 2000.

($F = 42.9$; df = 1,3; $P < 0.01$)

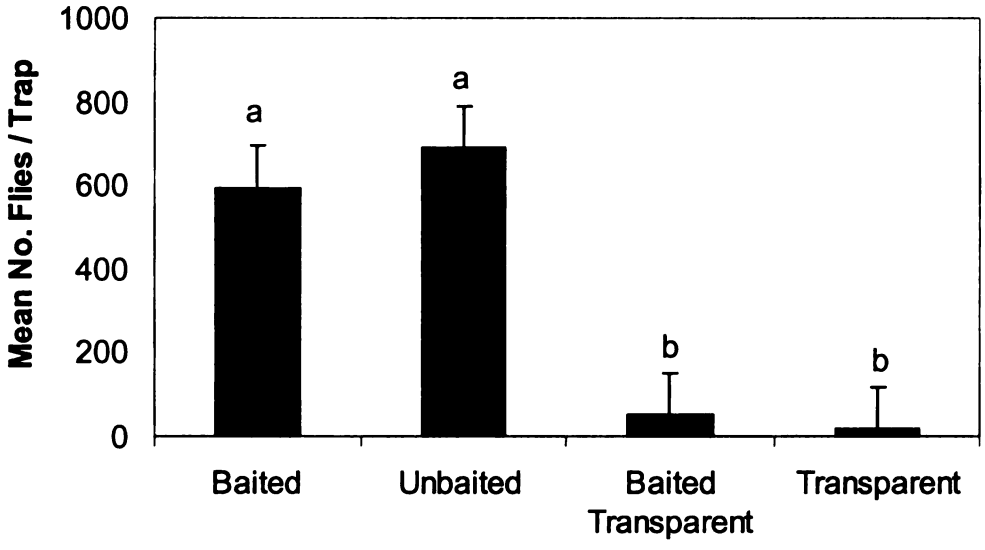

Fig. 4. Total captures of *Rhagoletis fausta* 24 May – 4 July on four types of modified Rebell™ traps, Paw Paw, MI (2001).

($F = 12.5$; df = 3,9; $P < 0.01$)

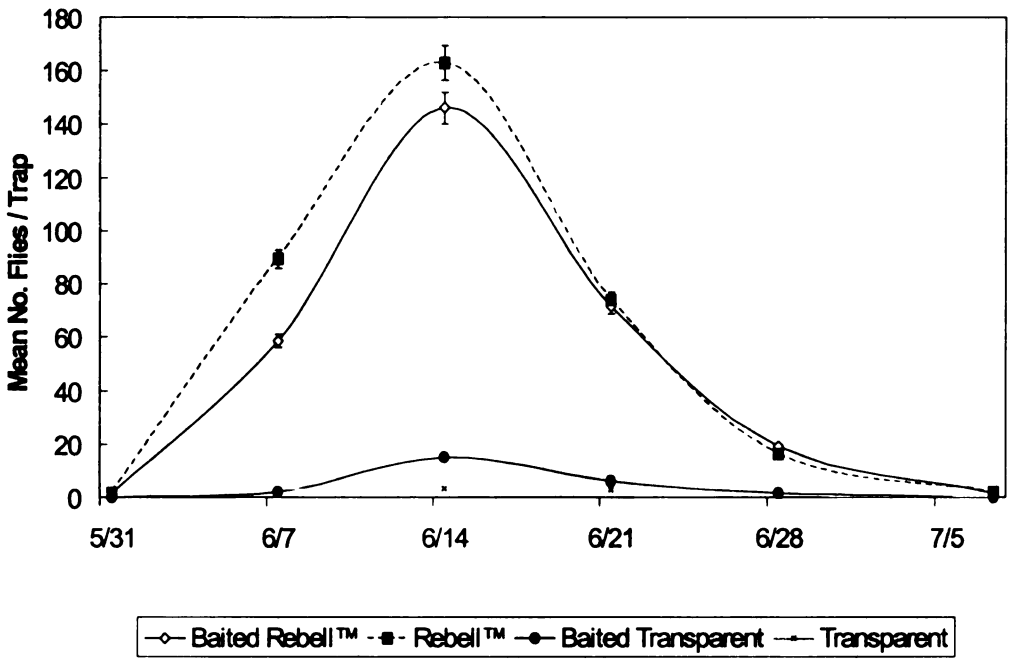

Fig. 5. Comparative population dynamics of *Rhagoletis fausta* as measured by four trapping systems in a cherry orchard, Paw Paw, MI 2001.

($F = 19.2$; df = 1,3; $P < 0.01$)

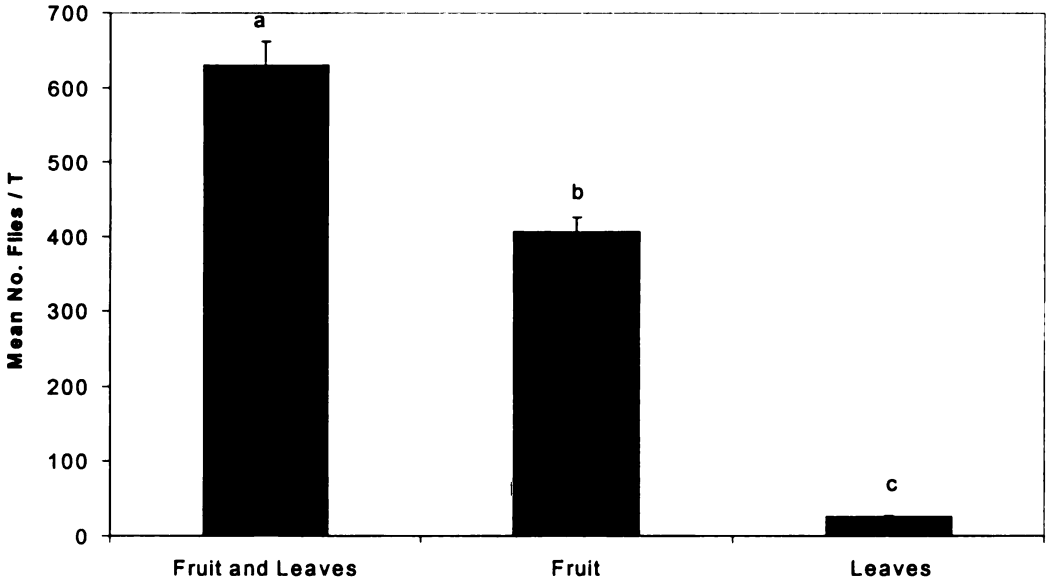

Fig. 6. Attraction of *Rhagoletis fausta* to Rebell™ traps in cherry trees, *Prunus cerasus,* Paw Paw, MI (2001).

CHAPTER TWO

EVALUATION OF REDUCED-RISK INSECTICIDES AS ALTERNATIVES TO ORGANOPHOSPHATE INSECTICIDES FOR CONTROL OF CHERRY FRUIT FLIES (DIPTERA: TEPHRITIDAE)

INTRODUCTION

The safety of pesticide application is a major concern for commercial cherry growers in Michigan. As a group, cherry growers in this region have been proactive, with the aid of research scientists at Michigan State University, in their approach to environmentally sound cultural and pesticide pest management practices. Michigan is currently the leading world-wide producer of tart cherries, *Prunus avium* L.

Approximately 75 percent of the nation's cherry production originates in Michigan, traditionally known as the "cherry capital of the world" (Michigan Agricultural Statistics 1999). To ensure pest and residue-free fruit to meet federally mandated zero tolerance levels, two to three applications of organophosphate insecticides are used in cherry orchards to control major economic pests, including the eastern, *Rhagoletis cingulata* (Loew) and black, *R. fausta* (Osten Sacken) cherry fruit flies (Edson *et al.,* 1998). Cherry fruit flies pose a threat to commercial cherry production in Michigan, in terms of domestic fruit quality and in the states share of the national cherry market. A major concern with organophosphates (OP's) used for pest control is that they kill not only pest organisms, but also non-target organisms, thus indiscriminately harming our natural resources.

Pest management is an integral part of agriculture. It brings world wide benefits to people, increasing both the quantity and quality of crops grown. Pesticides are commonly used in pest management. Their benefits obviously need to be delivered without posing unacceptable risks to non-target organisms. Thus, the potential risks, which are a function of both toxicity and exposure, need to be determined to ensure

pesticide safety. The 16th Century Swiss Physician, Paracelsus, said that it is "the dose that makes the poison". That exposure is a crucial element of risk was understood well before the modern advent of pesticides. Potential risks, however, are sometimes assessed and managed more by ranking their degree of toxicity, particularly where environmental fate data are lacking.

The discovery, development, registration, and commercial introduction of new pesticides for control of insect pests including cherry fruit flies, is a lengthy and expensive process. A significant amount of screening and targeted molecular design occurs in the identification of patentable chemistries with novel modes of action. Once a decision is taken to develop a new active ingredient, a major testing phase focused on field performance, health and environmental safety, manufacturing optimization, and commercial planning is initiated. This may last from three to five years, after which time a regulatory review period of anywhere from one-and-a half to four or more years occurs.

For every new active ingredient that enters development, there may be 20,000 or more candidate compounds and analogues that will have been screened for biological activity. Discovery and development phases are collectively both costly and time-consuming, and it may take five to ten years after entry into the marketplace for the original investment to be recovered. The cost of bringing a single product to the marketplace from discovery through launch may be $50 to 60 million or more. Thus, even a single year delay in reaching the marketplace can be very costly in terms of value recovery and patent protection. By using discounted cash flow analysis, it has been estimated that a one-year delay in registration approval and product launch may reduce the value of a new pesticide product by $5 million (Leng 1991).

Regulatory trends during the past decade or more have focused an inordinate degree of attention and an increasing level of resources on re-evaluation of older pesticides. Both technical and societal issues have driven the ever-increasing levels of regulatory scrutiny and increased pesticide registration requirements. Recent examples include interest in potential endocrine effects, acute dietary intake assessment, and ground and surface water contamination. In addition, political factors have yielded increased demands on the pesticide registration process. An example of this in the U.S. is the Clinton Administration's "Children's Health Initiative", which has driven highly conservative and restrictive approaches for pesticide evaluation despite lack of compelling evidence that children in the U.S. are at any undue risk of adverse effects from the use of pesticides (Huebner and Chilton 1998). The result of such increased scrutiny, which often has been generated by issues related to older products, is that it has become more difficult and expensive to register new products. The Environmental Protection Agency (EPA) is currently focusing on reducing pesticide use as a national priority. Furthermore, authorities have also been involved in comprehensive re-evaluation or re-registration programs focused on older chemicals. These programs are designed to review the safety aspects of existing pesticides in light of current data requirements and stringent assessment procedures.

Michigan cherry growers have strived to reduce use of insecticides to control cherry fruit fly through the implementation of alternative control strategies. Although this effort has brought some success, the pest management systems still relies primarily on organophosphate insecticides. Michigan cherry growers need economically viable and ecologically sound alternatives to organophosphate insecticides that will effectively

control cherry fruit fly and meet the stringent quality demands of the marketplace and government regulations. Zero tolerance is an extremely stringent standard. Even if growers continue to use organophosphate insecticides, there is a limit as to how far they can realistically (economically) reduce use, based solely on requirements that no cherry fruit fly larva be present in the fruit at harvest.

Current research trends have led to the findings and development of novel "reduced-risk" chemistries. The general principle of EPA's pesticide registration program in the U.S. is to give registration priority and accelerated approval to products with the most favorable characteristics (i.e. reduced-risk) as compared to conventional chemistry alternatives. The overall objective is to accelerate the introduction of reduced-risk products so that marketplace choices rather than increased regulatory restrictions can lead to replacement of older products and technologies. Under the program, pesticides classified as reduced-risk products must meet several or all of the following criteria as compared with currently available alternatives (U.S. EPA 1997): a) reduced risks to human health and non-target organisms; b) reduced potential for contamination of valued environmental resources (water, air, soil); c) broadened adoption of Integrated Pest Management (IPM). Reduced risk pesticide programs were first envisioned during the late 1980's and early 1990's. In 1992, public comments were invited on a proposed program for providing regulatory incentives for development of safer pesticide products. During 1993, a voluntary reduced risk pesticide initiative was introduced which described interim criteria for reduced-risk active ingredients, guidelines for submission of reduced risk rationale by registrants, and a streamlined registration process. By 1997, more detailed guidelines were promulgated for expedited regulatory review under the reduced-

risk program (U.S. EPA 1997), including adoption of formalized reduced risk criteria, a standardized submission format, and establishment of a Reduced Risk Committee at the EPA. During 1998, the EPA established a priority system of review for all registration submissions, with several categories of reduced-risk products adopted by the Agency as top priorities for registration review. In addition to first-time registrations of new active ingredients, the program was expanded in 1996 to include accelerated registration approval for additional crops and uses for active ingredients already classified as reduced risk.

As Food Quality Protection Act (FQPA) regulations lead to a reduction in the use of organophosphate insecticides, and public pressure against the use of broad-spectrum insecticides increases, it becomes necessary to identify effective non-organophosphate compounds for inclusion into novel pest management tactics. The objective of this study was to investigate the efficacy of novel chemistries as control alternatives to organophosphates (i.e. Guthion) for residential cherry fruit fly populations within orchard systems.

MATERIALS AND METHODS

Insecticide Trials. 2000. Experiments to evaluate the effectiveness of different kinds of insecticides on cherry fruit fly species were conducted in an abandoned sweet cherry (*P. avium)* block in Leelanau County, Michigan. The experiment began on 1 July, and was terminated on 25 August 2000. The experimental site consisted of a 2.05-acre block of non-sprayed cherries. The site has a dominant residential population of *R. cingulata* flies.

The experimental design consisted of a completely randomized block with four replicates. Plot size was approximately 3200 sq. ft. consisting of 5 x 5 trees (25 plants) spaced approximately 10 m within rows and 20 m between blocks. Fly populations were monitored by using baited Rebell™ traps (Swiss Federal Research Station, Wadswill, Switzerland). Baited traps had a green polycon dispenser containing 5.0 g of ammonium acetate attached to the top surface of the trap. The foliage immediately surrounding the traps was cleared to prevent any interference between the hung traps and the tree canopy (Drummond *et al.*, 1984). Insecticide applications were applied after detection of fruit fly emergence. Seven treatments were applied with a Friend Airblast Sprayer (Air-O-Fan, Reedley, California). Fly emergence was detected on 22 June 2000 (Fig. 7). The first application was made on 28 June 2000, and a second was made on 8 July 2000: 1) the Naturalyte, SpinTor® 2 SC (spinosad; 22.8% a.i.; Dow AgroSciences, Indianapolis, IN) at a rate of 6.0 oz/acre; 2) SpinTor Bait® GF 120 (0.02% a.i. bait) 52.0 oz./acre; three compounds of the neonicotinoid class, including: 3) Provado® 1.6 F (imidacloprid; 17.4% a.i.; Bayer, Kansas City, Missouri) at a rate of 8.0 fl oz/acre, 4) Calypso® 480 SC

(thiacloprid; 40.4% a.i.; Bayer Corp.) at a rate of 3.0 fl oz/acre, and 5) Actara® 25 WG at a rate of 4.5 oz/acre; 6) the conventional organophosphate, azinophos-methyl (Guthion® 50 WP (azinphos-methyl; 50% a.i.; Bayer Corp.) at a rate of 1.5 lbs/acre; and 7) an untreated control. An organo-silicant, Slygard® 309 (Diatect International Inc., Smith Center, Kansas) was added to SpinTor® 2 SC and SpinTor Bait® at a rate of 40 L / ha. The addition of Slygard® 309 allowed for a more uniform distribution of spray droplets on cherry trees. Experimental plot was sprayed with a fungicide, consequently no disease was detected.

Monitoring. Each Rebell™ trap was monitored for *R. cingulata*, and flies were removed twice per week. Female flies were classified as sexually mature or immature by thorough examination of the rear abdominal segments. Fruit infestation levels were monitored by collecting 200 cherries per treatment. They were placed over 0.5-cm hardware-mesh screens with containers below. Cherries were left on hardware mesh for three weeks at room temperature. Containers were examined on a daily basis for maggot emergence.

Laboratory Bioassays. 2001. Bioassays were conducted at our laboratory in the Center for Integrated Plant Systems at Michigan State University, East Lansing, Michigan. Single-Dose assays were conducted to study the behavioral response of sexually mature female flies (*R. fausta*) to selective compounds, and the conventional organophosphate control method, azinophos-methyl.

Fly Preparation. Cherries were collected from unsprayed farms in southwestern Michigan during the summer of 2000 and placed on 0.5-cm mesh hardware cloth (ACE Hardware, East Lansing, MI) for 21 days over collecting trays containing vermiculite (Liburd *et al.,* 1998b). Emerging puparia were collected from trays daily during the 21 day period and maintained at 5° C for approximately 9 months. Approximately 50 days prior to the start of the bioassays, puparia were placed in 60 x 60 x 60 cm stainless steel collapsible insect cages (BioQuip, Gardena, California) in shallow (2-cm deep) plastic containers with moist vermiculite. Puparia were kept moist by spraying water with an atomizer at 24° C and exposed to day length of 16 light:8 dark hours and 70% RH. After 35 days, cherry fruit flies began to emerge. Upon exclusion, newly emerged flies were provided with water and strips of cardboard coated with a mixture of yeast hydrolysate (enzymated autolyzed Brewer's yeast; 1 CN Biomedicals Inc., Costa Mesa, California), water, and honey. All flies, *R. fausta*, used in assays were tested when they were 10 days old because by that time they were believed to have mated and be reproductively mature (Hu and Prokopy 1998).

Single-Dose Response Assay. The following six treatment rates were used: 1) Naturalyte insecticide, SpinTor® 2 SC at a rate of 100 µl in 200 ml distilled water; 2) SpinTor Bait® at a rate of 1ml in 2ml distilled water; 3) Provado® 1.6 F at a rate of 100ul / 160 ml distilled water; 4) Calypso® 480 SC at a rate of 100ul / 400ml distilled water; 4) conventional organophosphate, Guthion 50 WP applied at a rate of 1.0 g / 400 ml distilled water; 6) untreated control. All treatments were mixed with SUN Ultra Fine Oil surfactant (Sun Refining and Marketing Co., Philadelphia, Pennsylvania) at 1% v/v

and administered at rates equivalent to field application by proportionate scaling of large quantity recipes.

Cherry cuttings from sour cherries with immature (green) fruit were obtained from an abandoned orchard in Paw Paw, Michigan in May 2001. Cherry shoots were homogeneous among replicates with 10 fruit of similar ripeness and repeatable foliage densities. Shoots were placed in transparent food containers (946 ml) with lids (SOLO CUP Company, Urbana, Illinois) were held turgid with moistened Oasis Floral Foam (Hyacinth House Greenery, Lansing, Michigan). Four flies per treatment (one fly per replicate) were introduced into the bioassay chambers. Food was provided in each chamber, and consisted of a mixture of yeast hydrolysate, honey, and water. Pin-sized holes were made in the lids of each container to allow fresh air to enter bioassay chambers. Cherry fruit flies were released into chambers and allowed to feed. Feeding duration was defined as time cherry fruit flies spent alighting on cherry cuttings while exhibiting proboscis extension. Observations to determine mortality were made during the 10-minute feeding bout, as well as 1 and 24 hours after feeding.

Statistical Analysis. Data from all experiments were square-root transformed ($x + 0.5$) and then subjected to an analysis of variance (SAS Institute 1989). Least Significance Test (LSD) was used to identify differences in treatment means ($P \leq 0.05$).

RESULTS

Insecticide Trials. 2000. On July 18, fruit infestation levels were significantly *($F = 11.3$; df = 6,9; $P < 0.001$)* greater in the untreated control plots compared with the rest of the treatments (Fig. 8). Fruit taken from plots treated with Actara®, Provado®, Calypso®, and Guthion® had 7 times fewer maggots than the untreated control. Plots treated with SpinTor® and SpinTor Bait® had 3 times fewer maggots than the control, these plots had significantly more maggots than Actara®, Provado®, Calypso®, and Guthion®. There was no significant difference between SpinTor® and SpinTor Bait®. Our final fruit sample on July 28 showed significantly *($F = 6.31$; df = 6,9; $P < 0.01$)* more maggots in the untreated control plots compared with the other treatments (Fig. 9). Apart from the control, there was no significant difference among the other treatments.

SpinTor®, however, had interesting results. In the first fruit analysis, there were significantly more larvae than both neonicotinoid and organophosphate treated plots. However, in the second fruit analysis (July 28) no significant difference was detected between SpinTor® and both neonicotinoid and organophosphate treated plots.

Laboratory Bioassays. 2001. After 10 minutes of feeding, significantly ($F = 10.4$; df = 5,8; $P < 0.001$) more cherry fruit flies were killed by fruit clusters treated with Calypso®, Provado®, and Guthion®; compared with SpinTor® and SpinTor Bait®. Twenty-four hours after the initial ten-minute feeding period, 100 % mortality was achieved among all chamber treatments (Fig. 10). No mortality was observed in the

flies that alighted and fed upon the untreated control chambers 24-hours after exposure (Fig. 10).

DISCUSSION

Experimental findings suggest that the neonicotinoid insecticides tested may be effective alternatives to conventional organophosphates for control of *Rhagoletis cingulata.* Fruit infestation analysis demonstrated that neonicotinoid insecticides were as efficient as conventional (organophosphate) control methods, though pest pressure was low to moderate. The two Spinosad formulations also were promising, though higher rates or more sprays may be needed to achieve enhanced control.

Laboratory bioassays provided similar data to those obtained in the field and helped explain some of the differences and changes in effectiveness that were observed over the course of the growing season. The assays showed that neonicotinoid insecticides were second behind Guthion® in killing fruit flies, with 100% mortality reached after 1 hour of exposure. Neonicotinoids were an effective means of controlling cherry fruit flies, though more time was needed to attain lethal activity compared to Guthion®. The naturalyte compounds (SpinTor® and SpinTor Bait®), were effective as well, but required substantially more time to kill than the neonicotinoids, thus indicating a positive correlation between field and laboratory trials.

These results are promising alternatives to organophosphates. While many insecticides are under review, and zero tolerance levels for maggot infested fruit remain, it is imperative to continue research on alternatives to control cherry fruit flies.

The reduced-risk program is by no means a panacea. Not all reduced-risk pesticides, having been afforded regulatory priority of effort, have become successful commercial products. In addition, not all reduced-risk products will necessarily pose less risk than current alternatives for all use patterns and in all circumstances. The concept of

reduced risk involves a relative comparison; an active ingredient may be considered reduced risk for some uses but not for others. Finally, the fact that a new or existing product has not achieved reduced-risk product status should not be construed as an indication that these types of products necessarily pose greater risks. The actual risk posed by the use of any pesticide product, whether it has been approved as a reduced-risk product or not, is a function of many factors including formulation type, method of application, use of protective clothing, and environmental variables.

We live in a world of choices when it comes to establishing the priorities of individual regulatory processes. As has generally been the case, massive investment of resources can continue to be devoted to cycles of data evaluation, regulatory re-evaluation and re-registration, and imposition of increased restrictions for the many older products currently employed in pest management programs. Some measure of attention to these older products is obviously warranted. However, with the continued limitation of resources in both the government and industrial sectors, an undue emphasis on the old may not allow sufficient resources to be directed to support the timely approval and market availability of the next generations of pesticides. The accelerated review and approval of new products with enhanced safety and environmental profiles is a proactive and positive step which will hopefully unleash marketplace forces to accelerate the replacement of older pesticide products. It is also an endeavour that places regulatory agencies and manufacturers in a collaboration which is of benefit to both pesticide users and detractors. Where we place our emphasis and energy in coming days, on the old or the new, will determine to a great extent how soon the world may have unfettered access to the safest and most desirable pest management tools.

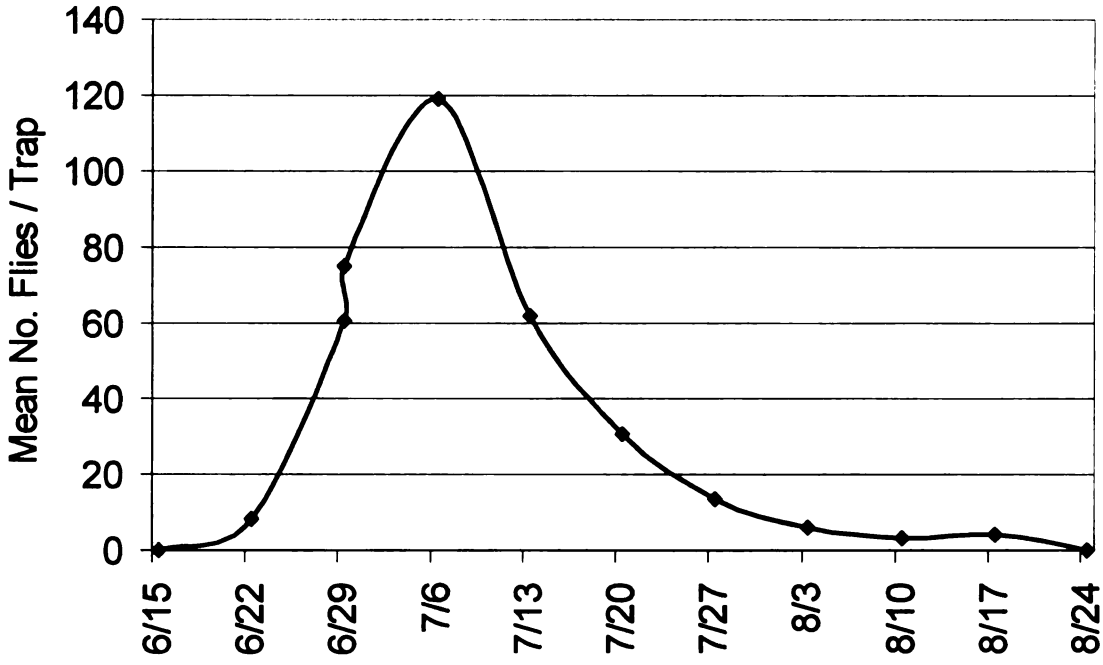

Fig. 7. Population dynamics of *Rhagoletis cingulata* caught on baited Rebell™ traps in an abandoned cherry orchard in Leelanau County, MI (2000).

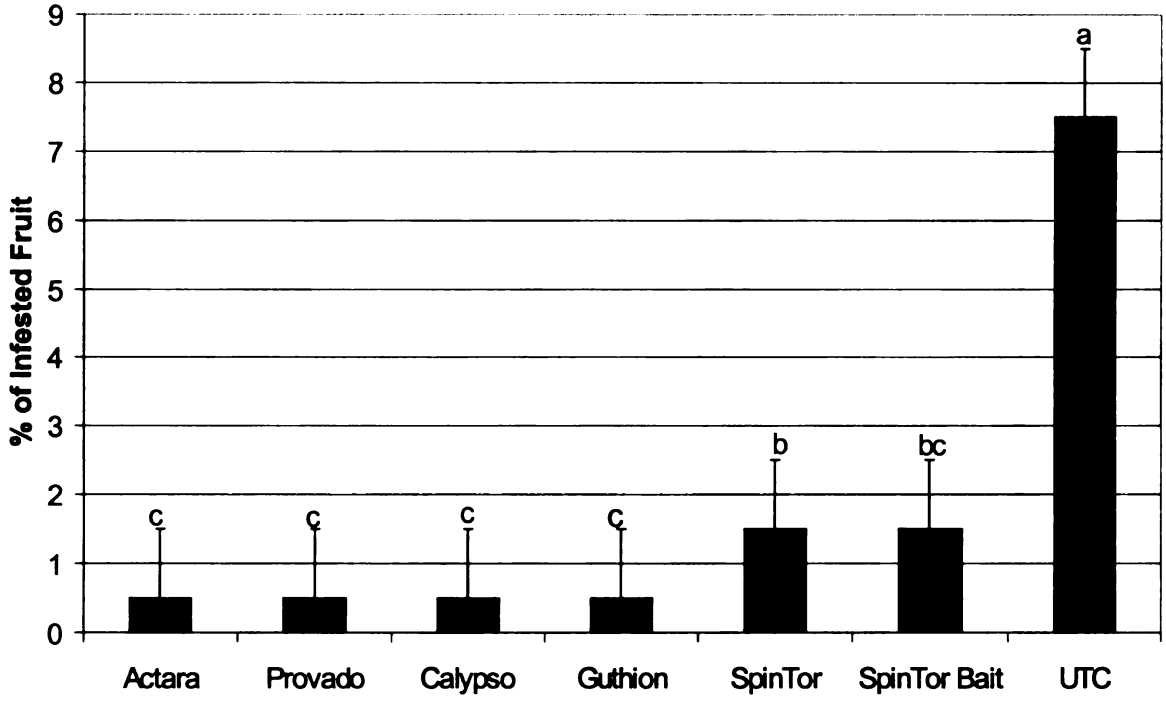

Fig. 8. Influence of six insecticides on *Rhagoletis cingulata* infestation of cherry fruit as measured on 18 July 2000 in a cherry research plot in Leelanau County, MI.

($F = 11.3$; df = 6,9; $P < 0.001$)

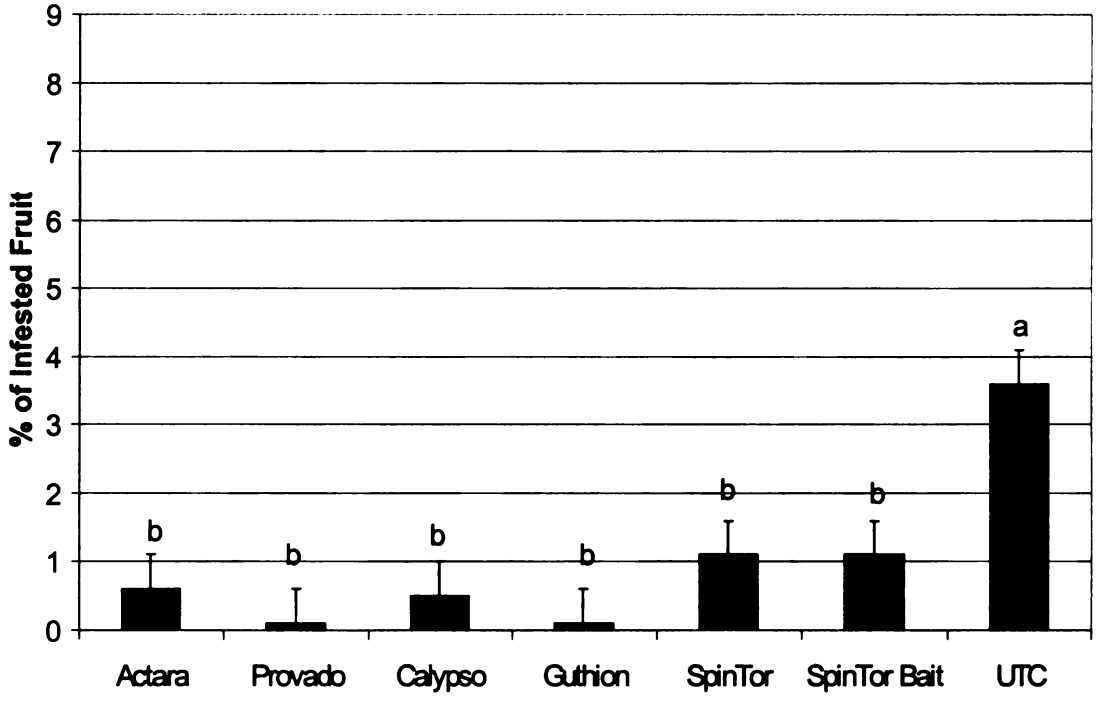

Fig. 9. Influence of six insecticides on *Rhagoletis cingulata* infestation of cherry fruit as measured on 28 July 2000 in a cherry research plot in Leelanau County, MI. ($F = 6.31$; df = 6,9; $P < 0.01$)

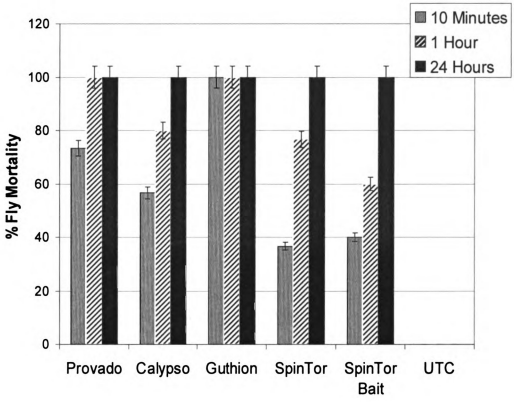

Fig. 10. Percent fly mortality (*Rhagoletis fausta*) exposed to different insecticides in bioassay chambers, Michigan State University, MI 2001.

CHAPTER THREE

EVALUATION OF BIODEGRADABLE, INSECTICIDE TREATED TRAPPING DEVICES AND ENTOMOPATHOGENIC NEMATODES FOR CONTROL OF CHERRY FRUIT FLIES, *RHAGOLETIS* Spp.

INTRODUCTION

The larval stages of two native tephritid flies feed on fruit in cultivated cherries are the eastern, *Rhagoletis cingulata* (Loew) and the black cherry fruit fly, *R. fausta* (Osten Saken). These have a major negative impact on the cherry industry. There is a zero tolerance among cherry growers, packers, consumers and international markets for cherries infested with maggots. Michigan Department of Agriculture inspectors constantly monitor packinghouses and sample cherry lots for cherry maggots. If a single maggot is detected, the packinghouse is shut down to dispose of all cherries that may be infested (Liburd *et al.*, 2001). This is often a lengthy and costly process.

To meet the zero tolerance for maggot-infested fruit, commercial growers rely on insecticides to protect their crop from cherry fruit flies (Edson *et al.*, 1998). Conventional broad-spectrum organophosphate insecticides are used. These are highly toxic chemicals and can threaten non-target organisms, particularly invertebrates in the immediate and surrounding habitats. Currently, most commercial cherry growers apply two to four foliar applications of organophosphate insecticides irrespective of the presence of flies (Liburd *et al.*, 1998).

As organophosphates are slowly phased out by Food Quality Protection Act (FQPA) regulations, a serious need arises for integration of multiple fruit fly control tactics in Michigan cherry production. The FQPA (1996) amends the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and the Federal Food, Drug, and Cosmetic Act (FFDCA). It alters the guidelines by which the Environmental Protection Agency (EPA) registers and regulates pesticide usage in the United States. This is accomplished by

mandating a health-based standard for essentially all foods. The standard is based on collective exposure to dietary and worker residue concerns. Moreover, it considers the effects of exposure to pesticides with common mechanisms of toxicity and demands a thorough screening for probable endocrine effects, especially when used on foods consumed by infants and children. Apples, peaches, pears and grapes are focal points for FQPA. Consequentially, EPA is re-registering and eliminating a significant number of pesticides under the requirements of FQPA. It has focused on organophosphates (OP) and carbamates (CB). These chemistries are traditionally used for fruit fly control in Michigan orchard systems. The potential restrictions are particularly important in relation to the zero tolerance for fruit flies associated with cherry marketing. There is currently a lack of alternative insecticides that are efficacious against this pest.

Integration of reduced-risk insecticide alternatives is crucial for a successful transition from current practices to the FQPA era. New insecticide chemistries are generally weak contact poisons. They produce, however, an array of sub-lethal effects such as oviposition deterrence. Neonicotinoids are an example of a new class of insecticides. With these materials, timing of the first cover spray is critical. It must coincide with cherry fruit fly egg-laying activities. Historically, growers have used yellow Pherocon AM board (Great Lakes IPM, Vestaburg, MI) to detect when the first fruit fly emerges. They make their first spray within a week of the trap catch (Prokopy 1975, Reissig 1976). Recent evidence, however, suggests that the three-dimensional Rebell™ trap (Swiss Federal Research Station, Wadswill, Switzerland) is significantly more efficient in detecting the initial flight of the cherry fruit fly following emergence

(Liburd *et al.,* 2001, Kostarides and Liburd *in press*). This trap caught more flies throughout the growing season, but also detected flies 1-2 wks earlier than the Pherocon AM board.

In an effort to reduce pesticide deposition into our environment, alternative methods of cherry fruit fly suppression are being researched. Recent studies have investigated the development of fruit-mimicking insecticide-treated spheres. These may be used as part of IPM programs. This could provide a potential alternative to broad-spectrum insecticide use for control of key *Rhagoletis* species. Laboratory and field studies have shown that significantly more of *Rhagoletis* sibling species, apple *R. pomonella* (Walsh) and blueberry, *R. mendax* Curran, maggot flies were killed with red and green biodegradable spheres treated with the insecticide imidacloprid (Merit® 75 WP, Bayer) compared with untreated controls (Hu *et al.,* 1998, Liburd *et al.,* 1999, Stelinski *et al.,* 2001). Mortality occurs after flies land on a trap and consume a lethal dose of the insecticide (Stelinski *et al.*, 2001). In addition, season-long residual activity studies indicate a fly mortality rate of 80% from spheres treated with imidacloprid at a rate of 1.5% a.i. (Hu *et al.*, 1998). Despite their effectiveness, there have been problems with biodegradable insecticide-treated spheres. These include fungal growth on the traps and loss of mass due to rodent feeding. Consequently, our research is designed to test new prototypes of these biodegradable traps.

Several advantages may be achieved from using biodegradable pesticide-treated sphere technology. They include season-long monitoring and the potential to control fruit flies with a single sphere deployment tactic. This requires less labor compared with other control devices and monitoring systems. There is also a reduced risk of insecticide

residues in fruit. Studies conducted during the 1999 field season showed that placing spheres at a distance 10 m around the perimeter of blueberry bushes was effective in intercepting immigrants (*R. mendax*) moving into blueberry plantings (Stelinski and Liburd 2001). Fields with a residential fly population, however, experienced higher levels of maggot injury. The integration of perimeter biodegradable-treated trapping devices used in concert with entomopathogenic nematodes (applied within orchards with residential maggot population) may help to suppress cherry fruit flies and reduce maggot injury.

Entomopathogenic nematodes are roundworms associated with a bacterium able to parasite and kill a large number of insects. These beneficial nematodes are microscopic, non-segmented worms that are associated with a bacterium able to parasitize and kill a large number of insects that occur naturally in soil all around the world. Insect-parasitic nematodes possessing an optimal balance of biological attributes are entomopathogenic nematodes in the genera *Steinernema* and *Heterorhabditis*. Entomopathogenic nematodes are extraordinarily lethal to many important soil insect pests, yet are safe for non-target organisms. This high degree of safety means that unlike most chemical pesticides, nematode applications do not require extensive safety equipment; and re-entry intervals, residues, contamination and pollinators are not issues. A copiousness variety of different insect pests are susceptible to infection, yet no adverse effects have been demonstrated against non-targets in field conducted studies (Georgis *et al.* 1991).

Once nematodes are released, they seek out host insects and enter their prey through body openings and emit an endo-toxin that results in death of the host insect

within 48 hours. The nematodes reproduce and their offspring feed on the insect cadaver and may emerge to seek out new hosts. Thus, entomopathogenic nematodes are nematode-bacterium complexes. The nematode may appear as little more than a biological syringe for its bacterial partner, yet the relationship between these organisms is one of classical mutualism. Nematode growth and reproduction depend upon conditions established in the host cadaver by the bacterium. The bacteria (*Xenorhabdus* sp.for *Steinernematids,* and *Photorhabdus* sp. for *Heterorhabditids*) further contributes anti-immune proteins to assist the nematode in overcoming host defenses and anti-microbials that suppresses colonization of the cadaver by competing secondary invaders. Conversely, the bacterium lacks invasive powers and is dependent upon the nematode to locate and penetrate suitable hosts.

Studies on Tephritids have demonstrated the potential for control with entomopathogenic nematodes (Beavers and Calkins 1984, Lindegren 1990). Finney (1983) found nematodes to be potential controls for *Rhagoletis* spp., and improvements in nematode strains, production, and storage should make nematodes an even more viable control option for cherry fruit flies. An appropriate soil environment is necessary for effective use of entomopathogenic nematodes (Smitley *et al.*, 1992). Application technologies for effective use of entomopathogenic nematodes in orchards have recently been developed. The use of entomopathogenic nematodes will specifically control larval and pupal population thereby reduce infestation in fields. Perimeter trapping not only enhances the nematode control strategy, but also prevents immigrants from entering the orchard. The net effect is a set of pest management tools for control of cherry fruit flies.

The goal of this project was to test the efficacy of perimeter-oriented control techniques and residential population control techniques together for a comprehensive fruit fly management program.

The objectives of this study were to: 1) evaluate the effectiveness of biodegradable pesticide-treated trapping devices against the two key species of cherry fruit flies; 2) develop and compare an insecticide treated trap modeled after the Rebell™ trap to be used as a control device for cherry fruit flies; 3) preform a chemical analysis to determine the fate of the insecticide on the modeled Rebell™ trap; 4) investigate the potential of using entomopathogenic nematodes for larval and pupal control through laboratory bioassays.

MATERIALS AND METHODS

Field experiments were conducted at th e Northwest Horticultural Research Station located in Leelanau County, Michigan. No insecticide sprays were applied to cherry blocks during experimentation.The experimental design was a completely randomized block with five replicates.

Biodegradable Pesticide-Treated Sphere Experiment. 2000. Field experiments were used to determine the effectiveness of biodegradable spheres treated with a novel neonicotinoid insecticide imidacloprid [Provado® 1.6 F (17.4% v/v a.i.; Bayer, Kansas City, Missouri)]. Spheres were commercially prepared with the specifications described by Liburd *et al.*, (1999). Essentially, spheres consisted of a mixture of water (150 g), table sugar (360 g), high fructose corn syrup (330 g), pregelatinized corn flour, (630 g), cayenne pepper (14.7 g), and sorbic acid (1.5 g). The spheres are designed to be both biodegradable and unattractive to wildlife. Prior to field deployment, spheres were primarily brush-painted with a base coat of fluorescent yellow paint. The second coat applied to the spheres consisted of 13 ml Provado® 1.6 F and 100 ml paint. The last (tertiary) coat applied to spheres was 13 ml Provado® 1.6 F, 80 ml paint, and 20 ml sucrose solution (fly feeding stimulant). The latex paint acts as a residue-extending agent for the insecticide and creates a barrier to control the release of both sugar and insecticide (Uh *et al.*, 1998).

Two treatments were evaluated. The traps were placed approximately 20 m apart within trees and 20 m between blocks. Treatment 1 consisted of a biodegradable sphere treated with the insecticide Provado® 1.6 at 2% a.i. and placed within the canopy of

cherry trees. Treatment 2 had an identical biodegradable sphere without the insecticide imidacloprid. Both spheres were baited with a polyethylene vial containing 5.0 grams of ammonium acetate (Liburd *et al.*, 1999).

Modified Pesticide Rebell™ Trap Experiment. 2001. Four treatments were evaluated. The traps were placed approximately 20 m apart within trees and 20 m between blocks. Treatments were as follows: 1) pesticide-treated wooden Rebell™ trap; 2) pesticide-treated biodegradable sphere; 3) untreated wooden Rebell™ trap (control); 4) untreated biodegradable sphere (control). Wooden Rebell™ trap dimensions were the same as the commercially available plastic monitoring Rebell™ trap. Wooden Rebell™ were first prepped brush painted with 200 ml "Karo" corn syrup mixed with 1qt of Glidden grey primer (Cleveland, Ohio). Two coats were applied.

Modified wooden Rebell™ traps were then treated with imidacloprid at a rate of 4% a.i.. The pesticide-treated biodegradable spheres were prepared as previously discussed for the biodegradable pesticide-treated sphere experiment in 2000. The 4% a.i. insecticide-treated traps were prepared as follows: traps were primarily brush-painted with a base coat of fluorescent yellow paint alone. The second coat applied to the traps consisted of 26 ml imidacloprid and 87 ml paint. The last (tertiary) coat applied to spheres was 26 ml imidacloprid, 67 ml paint, and 20 ml sucrose solution. Treatments 3 and 4 (untreated controls) were a modified wooden Rebell™ trap and biodegradable sphere, respectively, with no insecticide incorporated into the paint mixture.

Monitoring. Both traps and plexiglas panes was checked twice per week and the number of cherry fruit flies were counted, sexed, and removed.

Statistical Analysis. Data from biodegradable trapping device experiments were anaylzed by ANOVA followed by mean separation using the least significant difference test (LSD) test (SAS Institute 1989).

Entomopathogenic Nematode Bioassays. 2000. This study was conducted to determine the susceptibility of mature third instar larvae of *R. cingulata* to five species of entomopathogentic nematodes: *Steinernema feltiae* (Filipjev), *S. carpocapasae* (Weiser), *S. riobravis* (Cabanillas, Poinar, and Raulston), *Heterorhabditis bacteriophora* (Poinar), and *H. marelatus* (Liu and Berry).

Third instar larvae were obtained after emerging from field-collected unsprayed cherries. Infested cherries were placed on a wire screen and emerging larvae collected into a plastic pan with distilled water. Once removed from the water, larvae become active and pupated. Fruit fly larvae were exposed to an equivalent of a $10^6/m^2$ concentration of infective juvenile nematodes in petri plates (100 x 15 mm) lined with 9.0-cm-diameter Fisherbrand (P5) filter paper. Twenty larvae were selected randomly for each replicate plate. Distilled water (0.2 ml) was pipetted into all petri plates to moisten the filter paper. One milliliter of the appropriate nematode suspension (6360 infective juveniles/ml) was then added to each dish. Depending on availability of larvae, three to four replicate plates were used for each nematode species and the controls. Bioassays were repeated on two separate dates.

Due to the variability in availability of fruit fly larvae, the number of replicates was different for the two experiments. A distilled water control and four dishes per nematode species were used in Experiment 1. By the third week of July, when the second experiment was conducted, the availability of *R. cingulata* was limited. In the second experiment,

H. bacteriophora was not assayed and only three dishes per nematode species and control were used. Because pupation began soon after adding larvae to plates, an effort was made to select larvae in which pupation had not yet begun. Larvae which displayed distinctive mouth hooks, no brownish pigmentation, and no other signs of pupation were preferentially selected.

After addition of infective entomopathogenic juveniles, the petri dishes were covered, sealed with strips of parafilm, and incubated at 25°C. After 24 hours, the Petri dishes were unsealed and dry vermiculite was sprinkled over the pupae. The closed dishes were maintained at the same temperature and 55-60 % relative humidity for 7 days. Then 10 puparia were collected at random from each dish and dissected to determine pupal mortality and the presence of nematodes.

Living pupae were whitish-yellow with developing wing buds, wings, and legs. A distinct head, compound eyes, thorax, and abdomen were easily distinguished. Tissues of parasitized pupae were yellow-brown and contained infective juveniles and adult nematodes. Pupae were considered very little tissues, were yellow-brown without distinctive segmentation, or shriveled with remaining tissues fibrous. A visible tracheal system through the puparial shell indicated that the larva was infected by the nematodes

Statistical Analysis. Percentage mortality data of *R. cingulata* pupae were analyzed after square root arcsine transformation. Analysis and comparison of mortality data were done using ANOVA (General Linear Model procedure, SAS, 1996) followed by Least Significant Difference (LSD) test.

Pesticide Residual Analysis. This experiment was conducted to determine whether the concentration of insecticide used in wooden Rebell traps changed over the course of the growing season. Determination of total residues of the insecticide imidacloprid from pesticide-treated biodegradable spheres was done with extraction with an acidic aqueous solvent. The extract was filtered through a Celite vacuum filter flask and hydrophobic interferences were washed with hexane. The aqueous portion was retained and put onto a XAD4 cleanup column. The residues were eluted off the XAD4 column with methanol and oxidized to 6-chloronicotinic acid with potassium permanganate. Finally, the extract was analyzed with GC/MS.

Statistical Analysis. Data were subjected to analysis of variance (ANOVA) followed by Least Significant Difference (LSD) test.

RESULTS

Biodegradable Pesticide -Treated Trapping Devices. In the experiments to investigate the efficacy of biodegradable pesticide–treated spheres, significantly more *R. cingulata* were captured on traps with 2% a.i. compared with untreated spheres. Transparent plexiglas panes placed beneath treated spheres captured six times as many *R. cingulata* as the unbaited biodegradable traps (Fig. 11).

During the 2001 growing season, significantly ($F = 210.97$; df = 3,6; $P < 0.0001$) more *R. cingulata* were captured on wooden Rebell™ traps treated with 4% imidacloprid than any other traps tested (Fig. 12). Pesticide-treated wooden Rebell™ traps caught 6.8, 5.9, 2.0 times as many *R. cingulata* as the biodegradable pesticide-treated sphere, untreated wooden Rebell™ trap, and untreated biodegradable sphere. The biodegradable pesticide-treated sphere caught significantly more *R. cingulata* than both untreated traps (Fig. 12).

Entomopathogenic Nematode Bioassays. 2000. In Experiment 1, infestations of infective juveniles of *S. carpocapsae* and *S. feltiae* caused significantly higher pupal mortality in *R. cingulata* than did *S. riobravis* (Table 3). Sixty-five percent of the recently formed pupae were dead and contained *S. carpocapsae* juveniles.

Relative to the other four species, a greater number of *S. carpocapsae* infective juveniles entered the *R. cingulata* larvae, and juveniles were recorded in 31 of 40 pupae. Thirteen puparia each contained hundreds of *S. carpocapsae* infective juveniles.

Heterorhabditis bacteriophora killed a greater percentage of pupae then *S. feltiae*. Seven puparia were infected with *H. bacteriophora* and eight puparia with *S. feltiae*. Seven puparia of each species were infected with hundreds of infective juveniles. A greater percentage (50%) of pupae was dead but not infected with *S. feltiae*. Higher mortality of non-infected pupae may have been due to an interruption of metamorphosis.

Even though larval selection was randomized in Experiment 2, a higher mortality (43.3%) in the controls was observed. This may have been due to their collection later in the season, when lower numbers per cherry were found. Earlier in the season, adequate numbers of larvae were collected within a few hours of holding the cherries over water-filled trays. Later in the season it was necessary to hold cherries for up to 24 hours before adequate numbers of larvae could be obtained. Depending on when the larvae emerged from the cherries, they could have remained in water from 1 to 24 hours.

In the second experiment, larvae were also significantly more susceptible to infective juveniles of *S. carpocapsae* and *S. feltiae* than to *S. riobravis* (Table 3). *Steinermena carpocapsae* juveniles were found in 16 of 30 puparia, and 7 contained hundreds of infective juveniles. Ten of 30 puparia had *S. feltiae* juveniles and 11 puparia contained hundreds of juveniles. Sixty percent of the dead pupae had an *S. feltiae* infestation

(Table 3).

DISCUSSION

Biodegradable Trapping Devices. This study demonstrated that baited biodegradable trapping devices treated with the insecticide imidacloprid were more effective in killing cherry fruit flies than identical untreated devices. Specifically, baited biodegradable wooden Rebell™ traps treated with 4.0 % a.i. imidacloprid killed significantly more flies than all other traps tested.

These results show the potential for reduction of unnecessary spray depositions into our natural resources. Through improving upon trapping systems, we believe this will help facilitate the transition from conventional to alternative control technologies in Michigan fruit production systems.

Entomopathogenic Nematodes. *Steinernema carpocapsae*, because of their small size, may have infested more *R. cingulata* larvae than other species. This ambusher nematode is also active against other dipterans and had been adapted to infect insects at the soil-litter interface (Kaya and Gaugler 1993). When these nematodes are present in the soil, *R. cingulata* larvae could become infected as they drop onto the ground.

The use of entomopathogenic nematodes is really still in its infancy and there is considerable potential yet to be fulfilled (Bedding 1999). New species and strains of entomopathogenic nematodes are constantly being found and can now be stored in liquid nitrogen indefinitely to preserve genetic diversity (Popiel and Vasquez, 1991 Curran *et al.*, 1992). Most of our domestic animals and plants have been modified by artificial selection and this is undoubtedly possible with entomopathogenic nematodes

(particularly because their short life cycle) and has indeed been already attempted with some success. As better and better strains become available more kinds of insect pests can be targeted and fewer entomopathogenic nematodes will be required for treatments that will therefore become less expensive. There is also much research being conducted on methods for applying entomopathogenic nematodes that should help to further reduce treatment costs. However, in the end using entomopathogenic nematodes to control insects in orchard systems may be partly up to the ingenuity of the grower to find out the best possible means of and timing for applying them for their particular situation.

Table 3. Mortality of *Rhagoletis cingulata* pupae after exposure as larvae to $10^6/m^2$ concentration of infective juveniles of five species of entomopathogenic nematodes.

	Pupal Mortality	Nematode Recovery Infected (%)
Experiment 1		
S. carpocapsae	72.5 ± 1.27 a	65.0 ± 1.32 a
S. feltiae	70.0 ± 0.38 a	35.0 ± 0.50 bc
H. bactiophora	62.5 ± 0.79 ab	50.0 ± 0.41 ab
H. marelatus	55.0 ± 0.59 ab	15.0 ± 0.65 c
S. riobravis	40.0 ± 0.57 bc	17.5 ± 0.75 c
Control	22.5 ± 0.25 c	0.0 d
Experiment 2		
S. carpocapsae	83.3 ± 1.89 a	83.3 ± 0.67 a
S. feltiae	73.3 ± 1.26 a	60.0 ± 1.53 ab
H. marelatus	60.0 ± 0.93 ab	26.7 ± 1.45 bc
S. riobravis	40.0 ± 0.73 b	23.3 ± 1.33 bc
Control	43.3 ± 0.33 b	0.0 c

Means within same column followed by the same letter are not significantly different ($P \leq 0.05$; LSD test).

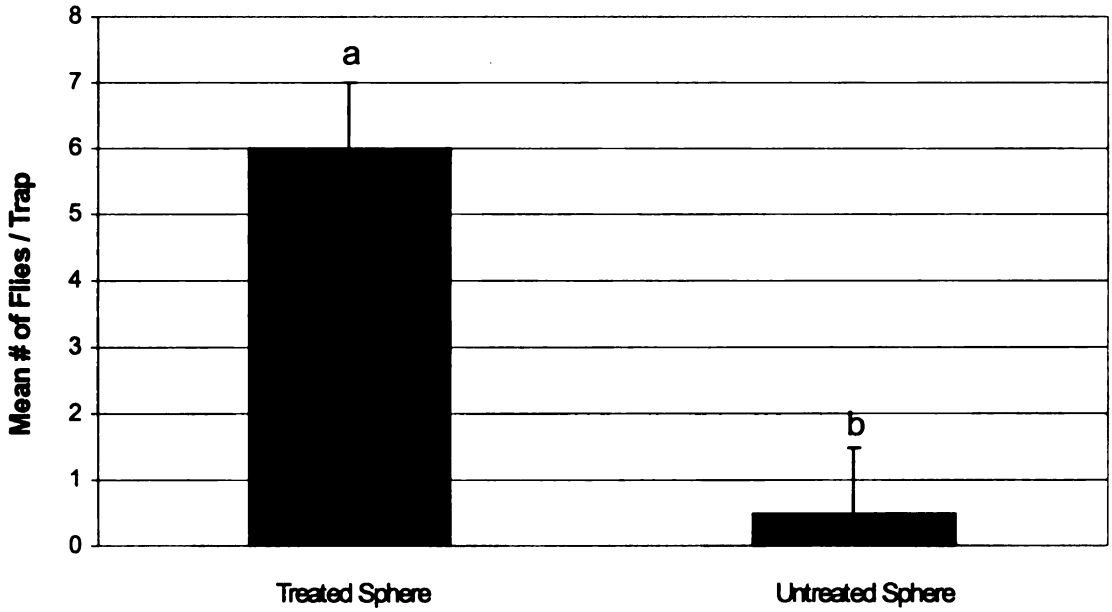

Fig. 11. Captures of *Rhagoletis cingulata* from 11-30 July captured on plexigals pans beneath biodegradable pesticide-treated (2% a.i. imidacloprid) spheres, Leelanau County, Michigan (2000).

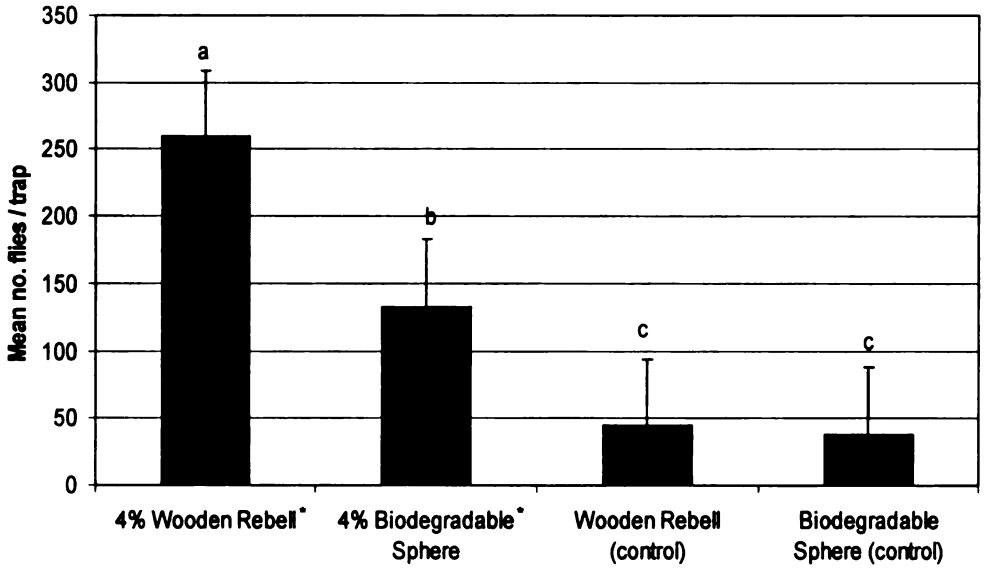

Fig.12. Captures of *Rhagoletis cingulata* from 1 July – 25 Aug. on plexiglas panels beneath biodegradable trapping devices, Leelanau County, Michigan (2001).

* 4% a.i. imidacloprid. ($F = 210.97$; df = 3,6; $P < 0.0001$)

SUMMARY AND CONCLUSIONS

Both visual (yellow color) and olfactory (ammonium baits) stimuli are important in the orientation of *R. cingulata* to Rebell™ traps. Therefore, I recommend that traps can be baited with ammonium compounds in order to effectively monitor for the eastern cherry fruit fly. In contrast, the visual stimuli appear to play a major role in the orientation of *R. fausta* to Rebell™ traps. Baiting traps with ammonium compounds, therefore, is not necessary. Furthermore, host resources that include fruit load and foliage affect the number of Rebell™ traps needed per hectare to effectively monitor cherry fruit fly populations.

The results from the evaluation of reduced-risk insecticides showed potential for future use of neonicotinoid chemistries in cherry fruit fly control. Fruit infestation levels indicated that these compounds were equally effective as conventional control methods in managing cherry fruit flies. Unlike organophosphates, the mode of action for neonicotinoids is host specific, thus not posing a threat to non-target organisms.

Biodegradable trapping devices treated with imidacloprid (Provado 1.6 F®) were effective in killing cherry fruit flies. Specifically, modifications of Rebell™ traps treated with 4% a.i. were highly attractive in luring adults to its surface and killing the flies. Based on these findings, I highly recommend deploying modified Rebell™ traps for both fly control and as a way to reduce pesticide deposition into the environment. To further decrease the number of insecticide applications needed in cherry orchards to suppress fly populations, entomopathogenic nematodes may have a role. Of the five species

evaluated, *Steinernema carpocapsae*, provided the best efficacy. Athough further research is advised for cost-effective application of entomopathogenic nematodes, laboratory assays have shown potential for their integration into a pest management system.

Overall, this research confirmed potential for use of Rebell™ traps for detecting the primary flight of residential fly populations. Upon emergence, it is advisable to deploy modified pesticide-treated (4% a.i. Provado® 1.6 F) Rebell™ traps to attract and kill cherry fruit flies. To further suppress pest populations, applying entomopathogenic nematodes to target fly larvae is an alternative to conventional sprays. The ability to develop a successful IPM program and minimize environmental risk that addresses the complexity of tropic interactions in agricultural systems holds the key to the future.

APPENDIX

APPENDIX 1

Record of Deposition of Voucher Specimens*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa, which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 2002-03

Title of thesis or dissertation (or other research projects):

Integrated Management Strategies For Control Of Cherry Fruit Flies, *Rhagoletis cingulata* and *Rhagoletis fausta* (Diptera: Tephritidae)

Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Investigator's Name(s) (typed)
Jessica Lynn Kostarides

Date 05/03/02

*Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24: 141-42.

Deposit as follows:

Original: Include as Appendix 1 in ribbon copy of thesis or dissertation.

Copies: Include as Appendix 1 in copies of thesis or dissertation.
Museum(s) files.
Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

Appendix 1

Table 4: Voucher specimen data

Page 1 of 1 Pages

Species or other taxon	Label data for specimens collected or used and deposited	Number of: Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Rhagoletis cingulata	MICHIGAN Leelanau Co. North West Hort. Station, MSU					10	10		MSU
Rhagoletis fausta	MICHIGAN Door Co. Abandoned Tart Cherry Orchard					10	10		MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Jessica Lynn Kostarides

Date 5/3/02

Voucher No. 2002-03

Received the above listed specimens for deposit in the Michigan State University Entomology Museum

Curator Date 5/3/02

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