1	Barry et al.: Organic Control of R. mendax	Address Correspondence to:
2	Journal of Economic Entomology	James D. Barry
3	Section: Horticultural Entomology	Blueberry & Cranberry Res. Center
4		125A Lake Oswego Road
- 5		Chatsworth, NJ 08019
6		Phone (609)726-1590 ext 12
7		Fax (609)726-1593
8		jamesbarry@aesop.rutgers.edu
9		
10		
11		
12		
13		
14	Comparative Effectiveness of Different Organic	Insecticides to Blueberry Maggot Flies
15	(Diptera: Tephri	tidae)
16		
17	JAMES D. BARRY, WILLIAM J. SCIARAPI	PA, ² SRIDHAR POLAVARAPU ³
18		
19	Blueberry and Cranberry Research and Exte	nsion Center, Rutgers University
20	125A Lake Oswego Road, Cha	atsworth, NJ 08019
21		
22	¹ E-mail: jamesbarry@aesop.rutgers.edu	
23	² Rutgers Cooperative Extension of Monmouth County	, Freehold, NJ
24	³ Author is recently deceased	d .

1 Abstract

2	Laboratory and field assays using organic insecticides were conducted on the blueberry
3	maggot, Rhagoletis mendax Curran. Direct contact of flies to Entrust® (spinosad), PyGanic®
4	(pyrethrum), and Aza-Direct® (neem extract) resulted in significantly higher mortality compared
5	to the control (water) after 2 and 24 h. After 24 h Entrust and Pyganic were not significantly
6	different from the positive control (phosmet); whereas Aza-Direct was significantly lower. A
7	second laboratory assay evaluated fly mortality to leaves 3 h after insecticide application. In this
8	assay, after 48 h PyGanic and Aza-Direct were not different from water, whereas Entrust
9	formulations were similar to the positive control (phosmet). The conclusion from these assays
10	was that Aza-Direct and PyGanic provided contact kill, but only Entrust had residual activity. A
11	repellency assay found no measurable effects for Aza-Direct. Large scale field trials found no
12	treatment affect for number of adult maggot flies captured in sticky traps; however, there were
13	significantly lower levels of fruit-infesting larvae in treated plots compared to the untreated
14	control. GF-120 NF Naturalyte Fruit Fly Bait® (spinosad bait), Entrust, and PyGanic were not
15	different from the positive control (imidacloprid). There was a significantly higher infestation in
16	the plot treated with Agroneem® (neem extract) compared to the positive control. The
17	implications for an organic integrated pest management program for blueberry maggot are
18	discussed.

21 KEYWORDS: Rhagoletis mendax, azadirachtin, pyrethrin, pyrethrum, spinosad

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1 Introduction

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The blueberry maggot fly, Rhagoletis mendax Curran, is a serious pest of lowbush and highbush blueberries, Vaccinium angustifolium Aiton and V. corymbosum L. respectively, in the northeastern United States and Atlantic Provinces of Canada (Geddes and Le Blanc 1987). It is often the most important insect pest of blueberries, because of the zero-tolerance for maggot presence in harvested fruit (Canadian Food Inspection Agency 1999). Although applying different types of insecticidal compounds, both organic and conventional farmers use either a calendar-based or an integrated pest management (IPM)-based spray program to control R. mendax. A calendar-based approach requires growers to start spraying insecticides within 10 d of the first detection of an adult fly in the area, and continue spraying at 7- to 10-d intervals until the end of harvest (Canadian Food Inspection Agency 1999). An IPM-based program requires growers to monitor the presence of adults using ammonium acetate baited sticky traps (Wood et al. 1983, Geddes et al. 1989). A recommended insecticide should be applied within 5 d of the date of capture of a single fly in any one of the monitoring traps, followed by a second spray 7-10 d later. This spray interval should be repeated for each subsequent fly detection until the end of harvest. Broad-spectrum insecticides, such as organophosphates and carbamates, have been the

Broad-spectrum insecticides, such as organophosphates and carbamates, have been the cornerstone for conventional control of *Rhagoletis* flies. In addition to these compounds being unavailable to organic growers, there was no universal definition on what was an acceptable organic pesticide. The creation of the USDA National Organic Program (NOP) in October 2002 and the Organic Materials Review Institute (OMRI) provided a framework to evaluate and certify compounds as organic that met a set of established standards.

Several different types of OMRI-registered insecticides have potential for providing 1 2 effective control of blueberry maggot. One group of organic insecticides is based on extracts from the neem tree (Azadirachta indica A. Juss.), some of which have repellant, feeding 3 inhibition, and anti-hormonal activity in insects (Nat et al. 1991). Azadirachtin is the primary 4 5 compound responsible for this activity in insects and Stark et al. (1990) found azadirachtin to 6 affect metamorphosis, longevity, and reproduction in several tropical tephritids. However, Prokopy and Powers (1995) found no affect on oviposition or mortality of R. pomonella (Walsh). 7 8 There are currently 14 different organic formulations of this insecticide available. 9 A second type of insecticides contain spinosad, which was developed from the bacterium 10 Saccharopolyspora spinosa Mertx and Yao. There are organic formulations of this toxicant as a 11 bait spray and a spray. Bait sprays, which contain ammonia-based attractants, a feeding 12 stimulant such as sucrose, and an insecticide, have been used to control outbreaks of Anastrepha 13 ludens, Bactrocera dorsalis (Hendel), and Ceratitis capitata since the 1950's in the United States (Steiner 1952, Moreno and Mangan 2003). The bait that is now commercially used with 14 15 spinosad, GF-120 NF Naturalyte Fruit Fly Bait, was based on SolBait, which was developed by 16 Moreno and Mangan (2003). Barry and Polavarapu (2004) found SolBait to be the best of 17 several baits tested in evaluations on R. mendax. Non-organic formulations of bait sprays 18 containing spinosad were developed for controlling tropical and sub-tropical tephritid pests, with 19 feeding studies confirming high mortality for Anastrepha ludens (Prokopy et al. 2000), A. 20 suspensa (King and Hennessey 1996), Bactrocera cucurbitae (Prokopy et al. 2003), and 21 Ceratitis capitata (Peck and McOuate 2000, Vargas et al. 2001, Barry et al. 2003). 22 The last group contain pyrethrum, an insecticide produced primarily in the flowers of 23 Tanacetum cinerariaefolium L. Pyrethrum has good knockdown ability, but very short residual

activity, as it degrades rapidly after exposure to sunlight (Katsuda 1999). Many synthetic 2 insecticides based on pyrethrum chemistry, called pyrethroids, have been developed to increase the residual activity. 4 In addition to insecticide sprays, previous work by Prokopy (1975) found painted 5 spheres, baited with ammonia compounds, to be highly attractive to tephritids. Spheres were first coated with a sticky material to trap flies (Prokopy 1975), but the need for decreased 6 deployment and handling time necessitated finding an insecticide replacement (Duan and 7 8 Prokopy 1995). Several studies have found spheres to be effective in controlling R. mendax and R. pomonella (Duan and Prokopy 1995, Liburd et al. 1999, Ayyappath et al. 2000, Stelinski et al. 10 2001). However at present the use of spheres is not commercially economical for either conventional or organic blueberry growers, because of the necessary deployment density and the 12 associated costs of products (i.e., spheres and residue extending agents) and labor (i.e., monitoring and applying insecticides to spheres) (Barry and Polavarapu in press). Our goal was to identify the most effective organic insecticide for controlling R. mendax 14 15 populations in highbush blueberry. 17 Materials and Methods

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Insects. Infested blueberries were collected near Chatsworth, NJ, in the summer and fall of 2003. The rearing procedures of Ayyappath et al. (2000) were used to obtain adult R. mendax for studies during 2004. Briefly, infested berries were placed over moist sand for larvae to drop and pupate. Puparia were sifted from sand three-five weeks later and kept in a screenhouse. Puparia were transferred to an incubator in November, at 6°C with a photoperiod of 12:12 (L:D) to complete diapause. The following March puparia were placed at 8°C. Periodically, groups of

puparia were transferred from 8 to 15°C for approximately 8 d and then transferred to an 1 incubator at 25°C with a photoperiod of 16:8 (L:D) until adult emergence, which occurred 25 to 2 45 d later. Adult flies were kept at 21-23°C and were provided a diet of sucrose and water. Flies 3 4 used in assays were 7-13 d-old and allowed to acclimatize to experimental conditions for several 5 hours before trials commenced. 6 Direct Contact with Fresh Insecticide. Fly survivorship was evaluated after direct 7 exposure to insecticides in 1 liter plastic containers with a screened top. Prior to exposure, ten 8 flies were placed in each container with a moist cotton ball. In addition to water and the 9 organophosphate phosmet (20.8 oz/ac; Imidan, Gowan Co., Yuma, AZ), there were two rates of 10 the following organic insecticides: Aza-Direct (16 and 32 fl oz/ac; Gowan Co), Entrust (1 and 2 oz/ac; Dow AgroSciences, Indiananopolis, IN), and PyGanic (16 and 32 fl oz/ac; McLaughlin 11 12 Gormley King Company, Minneapolis, MN). All treatments were prepared at a rate of 25 gallons of water per acre. A total of 8 replicates were completed for each treatment. Insecticides 13 14 were applied using a spray bottle (8-oz; Goody Products, Atlanta, GA) to mist approximately 0.7 ml per container through the screened top. Fly mortality was determined 2 and 24 h after 15 16 exposure. This experiment occurred in a screen house with temperatures ranging from 20-3017 °C. 18 Exposure to 3hr -old Insecticide. Fly survivorship was evaluated after exposure to 19 insecticides in 1 liter plastic containers with a screened top. Eight treatments (the same as in 20 Direct Contact with Fresh Insecticide section) were applied to each of eight blueberry bushes. 21 Three hours after application (when vegetation was dry) 3 leaves from each treated plant were 22 collected and placed in a 1 liter plastic container with a screened lid, which contained a moist cotton ball. Ten flies were then placed in each container. Fly mortality was determined 24 and 23

1 48 h after the start of exposure. This experiment occurred in the laboratory with temperatures 2 ranging from 21 - 23 °C. 3 Repellency. The repellency of Aza-Direct was assessed in a laboratory assay. Aza-Direct was prepared (32 oz/ac in 25 gallons of water/acre) and applied to foliage of a blueberry 4 5 plant until leaves were dripping. The control was a blueberry plant that was misted with (tap or 6 distilled?) water until dripping. Three hours after application (when vegetation was dry) 40 7 leaves from each treated plant were collected. Five flies were placed in a plastic container (12.5 8 cm in height and 13 cm in diameter) to which two leaves of one treatment were added. A total of 9 20 replicates were completed. Containers were observed for 70 min and any fly remaining on a 10 leaf for at least 3 s was removed. Observations were made in the laboratory at 21-23 °C. 11 Field Spray Trial. Four organic insecticides were evaluated, GF-120 NF Naturalyte 12 Fruit Fly Bait (20 fl oz/ac), PyGanic (32 fl oz/ac), AgroNeem (64 oz/ac; Agro Logistic Systems, 13 Diamond Bar, CA) and Entrust (2 oz/ac), in a field spray trial to assess blueberry maggot control. 14 This study was conducted in a recently abandoned blueberry field, located in Tinton Falls, NJ. 15 Blueberry bushes were approximately 2 m in height and spaced 1.2 m within and 2.7 m between 16 rows. Treatment plots were 30.5 m by 91.4 m, approximately 0.28 ha. Treatments were 17 replicated three times. 18 Each insecticide, except GF-120, was mixed with 25 gal of water/acre and sprayed using 19 a commercial (air blast) mist blower to both sides of bushes. GF-120 was mixed at a ratio of 1:5 20 with water for 120 oz of mix/acre, and applied to one side of every row using an ATV Olive 21 Fruit Fly Sprayer (PBM Supply & Mfg, Chico, CA). This ATV sprayer had a 25 gal tank, a non-22 adjustable 100 psi pump, and two nozzles from airblast sprayers consisting of discs (D-3 orifice) 23 without the core. Insecticides were applied on 7, 15, 22, and 29 Jul 2003. Adult blueberry

1 maggot fly presence was assessed twice a week for 6 weeks using four Pherocon AM traps baited with ammonium acetate (Great Lakes IPM, Vestaburg, MI) per plot. Larval presence was 2 determined on three dates (24 and 31 Jul, and 12 Aug) by randomly selecting 1,000 berries in 3 4 center rows from each plot and boiling fruit (Pickett and Spicer 1931) or Polk (microwave). 5 Maggots extracted from these samples were counted. Statistics. Fly mortality in direct and 3-hr old exposure assays, and flies landing on 6 leaves in the repellency assay were arcsine square-root transformed and analyzed using analyses 7 8 of variance (ANOVA) (SAS Institute 1999). Means were separated using Fisher's least 9 significant different (LSD) tests. Adult and larval numbers in field spray trial were square-root transformed ($\sqrt{x} + 0.5$) and arcsine square-root transformed, respectively prior to ANOVA and 10 11 means were separated using Fisher's LSD test. 12 13 14 Results Direct Exposure to Insecticide. Exposure of flies to insecticides resulted in significant 15 differences in fly mortality after 2 and 24 h (F = 74.58; df = 7, 56; P < 0.0001; and F = 49.2; df = 16 7, 56; P < 0.0001; respectively; Table 1). After 2 h fly mortality was significantly higher in all 17 insecticide treatments compared to the control, with greater than 90% control in Imidan, and high 18 19 and low rates of PyGanic. High and low rates of Entrust and Aza-Direct resulted in 20 approximately 50% mortality. After 24 h fly mortality approached 100% for all treatments 21 except rates of Aza-Direct, which were approximately 65%, and the control which was?. 22 Exposure to 3hr -old Insecticide. Treatments resulted in significant differences in fly mortality after 24 and 48 hours of exposure (F = 33.99; df = 7, 56; P < 0.0001; and F = 37.77; df 23

= 7, 56; P < 0.0001; respectively; Table 2). Fly mortality approached 40% for both rates of

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1	Entrust after 24 n and was close to 90% after 48 n, both significantly higher than the control.
2	After 24 h fly mortality was near 100% in the Imidan treatment and after 48 h, Imidan was not
3	significantly different from the high rate of Entrust. After 24 and 48 h neither rate of Aza-Direct
4	nor the high rate of PyGanic was significantly different from the control, which was
5	approximately 4 and 21%, respectively. The low rate of PyGanic was significantly different from
6	the control after 24, but not 48 h.
7	Repellency. Application of Aza-Direct to leaves did not result in a significant difference
8	in the number of flies landing on leaves ($F = 1.75$; df = 1, 38; $P = 0.194$). Numerically more
9	flies were removed from leaves in the control than Aza-Direct (31 vs. 21 percent, respectively).
10	Field Spray Trial. Treatments had no statistically significant affect on the number of
11	adults captured in traps (table 3) due to high variability within the check. Nonetheless, there was
12	a general trend towards reducing fly numbers from any of the treatments. The positive check,
13	Provado, had the least number of adults numerically (~15 flies per plot), followed by GF-120
14	and Entrust (~100), AgroNeem and PyGanic (~150), and the control (225) (Fig. 1). Treatments
15	had a significant affect on the percent of berries with maggots on each of the three sampling
. 16	dates (Table 4). Larval infestation in the untreated check plot was approximately 12% and was
17	significantly higher than any of the insecticide treatments (Table 4). The numbers of larvae in
18	the GF-120, Entrust, and PyGanic plots were not significantly different from Provado, whereas
19	the Agroneem plot was always significantly higher than Provado (Table 4).
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21	Discussion
22	All organic insecticides had some ability to control blueberry maggot, resulting in the
23	potential for an insecticide rotation schedule that minimizes resistance development. In both

1 laboratory assays fly mortality was initially highest in the organophosphate treatment, but after a period of 1-2 d fly mortality was comparable between the organophosphate and several organic 2 3 treatments. In this study, residual activity was tested at a maximum of 3 h after application. At a 4 larger interval, differences in residual activity between organic and organophosphate insecticides 5 are expected to manifest. 6 Aza-Direct and PyGanic resulted in fly mortality only near application time, whereas 7 spinosad-based compounds had a longer period of residual activity. The residual activity of 8 these organic insecticides is related to the PHI (pre-harvest interval). The pre-harvest interval is 9 3 d for Entrust and 0 d for Aza-Direct, PyGanic, and GF-120 NF Naturalyte Fruit Fly Bait. 10 Although Entrust applications resulted in the highest mortality of any organic insecticide in 11 laboratory assays, the PHI restriction could present a challenge to U-pick operations that are 12 open everyday. In our Field Spray Trial insecticides were all applied on either Monday or 13 Tuesday because the grower was open only on the weekend. Compounds with a 0 PHI allow 14 growers the option to spray the night before harvesting fruit. Although bait spray use requires obtaining specialized application equipment, promising 15 results were obtained in the field spray trial. It was not expected that spinosad spray and 16 17 spinosad bait spray would be comparable, because 400x less active ingredient was used in the 18 latter (Entrust: ai/ac = 80% of 2 oz = 1.6 oz; compared to GF-120: ai/ac = 0.02% of 20 oz = 19 0.004 oz). In a study with R. pomonella on apples (Malus sp.), GF-120 blank (without spinosad) 20 was found to increase maggot infestation in comparison to both a control and GF-120 (with 21 spinosad), suggesting that sufficient numbers of flies are capable of locating bait (K. Pelz, R. 22 Isaacs, J. Wise, L. Gut, Michigan State University, East Lansing, MI, unpubl. data). Further

study is necessary to determine the use of bait sprays as potential border sprays (Prokopy et al.

2 2003).

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3 There was at least one maggot found in each treatment on each of the three occasions that

4 berries were randomly collected and boiled., even in the non-organic control of imidacloprid.

5 Had our trials occurred in a commercial setting, instead of a previously abandoned field, the

maggot infestation rate would have been lower due to selective harvest. The pest pressure in our

trials was also increased because of the close proximity of a large area of unsprayed blueberries

(~10 acres), which provided a continual source of blueberry maggot migration into the

experimental plots.

An organic integrated pest management program for blueberry maggot is likely to involve multiple sprays to achieve sufficient control. To avoid resistance we recommend switching to a product with a different active ingredient after each spray. In the simplest rotation, sprays would alternate between two compounds (i.e., spinosad, neem extract, spinosad...). Three different active ingredients were evaluated in this study (neem extract, pyrethrum, and spinosad). (The active ingredient in Entrust and GF-120 is the same, so use of both would not be considered a rotation.) Specific decisions on what insecticide to spray could be impacted by the following factors: resident or local maggot population, maggot pressure, presence of other pests, days to harvest, and previous insecticide use.

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3	We thank Elizabeth Bender, Robert Holdcraft, Linda Tran-Barry, Jenn Hall, and Kate
4	Hall for assisting with laboratory work; John Applegate, Amy Testa and Luís Teixeira for
5	counting flies and Vivian Quinn, Pierre Tannous and the Monmouth County Master Gardeners
6	for collecting and processing berries; Agro Logistic Systems, Dow AgroSciences, Gowan, and
7	McLaughlin Gormley King for providing insecticides; and John Marchese and Wayne XXX for
8	applying insecticides and maintaining fields. This research was funded in part by SARE -
9	Sustainable Agriculture Research and Education Program grant ONE 03-016XXXX, Hatch
10	Project # 08189, and a grant from USDA - CSREES - Risk Avoidance and Mitigation Program
11	2002-51101-01903. This is New Jersey Agricultural Experiment Station Project # 31001.
12	
13	James – the name of the SARE grant is Ratcheting Up Commercial Organic Highbush Blueberry
14	Production Systems

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Table 1. Mortality from Direct Exposure to Organic Insecticides

		Mortality (%)		
Treatment	AI (oz/ac)	2 h	24 h	
Aza-Direct	16	$52.5 \pm 3.6c$	$62.5 \pm 4.9b$	
	32	$43.7 \pm 7.3c$	68.7 ± 8.9 b	
Entrust	1	$60.0 \pm 4.6c$	$100.0 \pm 0.0a$	
	2	$46.2 \pm 8.2c$	$100.0\pm0.0a$	
PyGanic	16	100.0 ± 0.0 b	100.0 ± 0.0 a	
	32	$93.7 \pm 2.6a$	$96.2 \pm 2.6a$	
Imidan ¹	20.8	$95.0 \pm 3.2ab$	100.0 ± 0.0 a	
Water (control)	an an an	$0.0 \pm 0.0 d$	21.8 ± 6.6 c	

Means in the same column having the same letter are not significantly different (Fisher's LSD, P = 0.05)

^{= 0.05})

I organophosphate insecticide was included as a positive check

Table 2. Mortality to 3 h-old Organic Insecticides

		Mortality (%)		
Treatment	AI (oz/ac)	24 h	48 h	
Aza-Direct	16	7.5 ± 3.1 cd	$32.5 \pm 5.9c$	
	32	8.7 ± 3.9 cd	$23.7 \pm 4.6c$	
Entrust	1	$38.7 \pm 7.6b$	$93.7 \pm 3.2ab$	
	2	$36.2\pm8.0b$	85.0 ± 5.9 b	
PyGanic	. 16	$10.0\pm2.6c$	$25.0 \pm 5.6c$	
	32	13.7 ± 3.7 cd	$37.5 \pm 7.0c$	
Imidan ¹	20.8	$96.2 \pm 1.8a$	$98.7 \pm 1.2a$	
Water (control)		$3.7 \pm 2.6d$	$21.2 \pm 5.8c$	

Means in the same column having the same letter are not significantly different (Fisher's LSD, P = 0.05)

1 organophosphate insecticide was included as a positive check

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Table 3. Adult captures in traps

Number of flies captured in 4 Pherocon traps $(\text{mean} \pm \text{SE})^1$						
Treatment	27 Jun – 6 Jul	7-15 Jul	16-21 Jul	22-28 Jul	29 Jul – 4 Aug	5-11 Aug
Entrust	47.0 ± 7.4	35.7 ± 6.7	15.0 ± 4.9	16.7 ± 2.7	7.7 ± 2.3	21.3 ± 7.1
GF-120	68.7 ± 26.6	25.0 ± 10.6	8.0 ± 2.3	16 ± 2.7	11.7 ± 5.2	30.0 ± 15.5
Agroneem	84.0 ± 15.7	44.0 ± 11.0	20.0 ± 11.2	15.7 ± 9.7	36.3 ± 17.5	23.7 ± 5.2
PyGanic	90.7 ± 45.4	40.0 ± 22.8	48.0 ± 40.7	28.0 ± 20.5	14.3 ± 8.8	14.7 ± 7.3
Provado ²	48.7 ± 16.1	5.7 ± 3.5	1.3 ± 0.3	1.7 ± 1.2	1.0 ± 0.6	4.7 ± 0.3
Untreated	40.0 ± 17.0	76 ± 41.8	50.7 ± 21.7	54.0 ± 30.0	23.7 ± 10.8	20.7 ± 10.1
F	0.58	0.49	1.22	2.06	2.29	2.54
P	0.717	0.781	0.36	0.142	0.111	0.086

df = 4, 10¹ insecticides applied on 7, 15, 22, and 29 Jul
² organophosphate insecticide was included as a positive check

Table 4. Larval presence in blueberry fruit

		Percent of boiled blueberries with maggots ¹			
Treatment	Rate (oz/ac)	24-Jul	31-Jul	12-Aug	
Entrust	2	0.57 ± 0.20 bc	1.00 ± 0.46 bc	0.67 ± 0.34 bc	
GF-120	20	$0.47 \pm 0.17 bc$	$0.53 \pm 0.03c$	0.53 ± 0.22 bc	
Agroneem	64	$2.27 \pm 1.48b$	$2.30 \pm 1.08b$	1.90 ± 1.25 b	
PyGanic	32	1.03 ± 0.34 bc	$0.70\pm0.23\mathrm{c}$	$0.50 \pm 0.06 bc$	
Provado ²	6	$0.3 \pm 0.06c$	$0.23 \pm 0.08c$	$0.03 \pm 0.03c$	
Untreated	san rute ma	$13.70 \pm 3.04a$	$14.10 \pm 4.50a$	$9.7 \pm 2.25a$	
F		19.49	25.29	13.55	
P		< 0.0001	< 0.0001	< 0.0001	

Means in the same column having the same letter are not significantly different (Fisher's LSD, P = 0.05)
¹ df = 5, 66

² organophosphate insecticide was included as a positive check

Figure Legends

Fig. 1. Number of adult blueberry maggot flies (mean \pm SE) captured for the season after the first insecticide spray. (NS, P > 0.05, ANOVA)

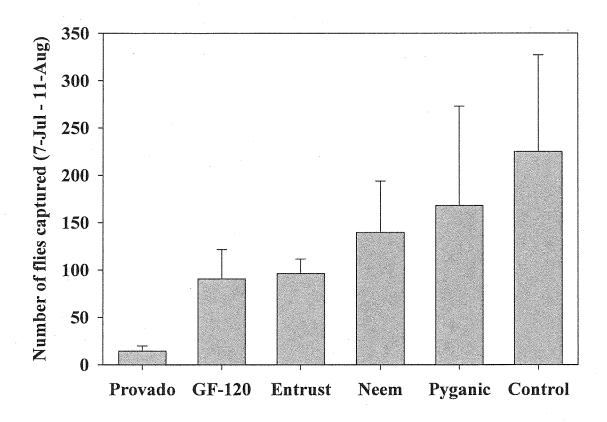


Fig. 1.