MORTALITY OF *RHAGOLETIS POMONELLA* (DIPTERA: TEPHRITIDAE) EXPOSED TO FIELD-AGED SPINETORAM, GF-120, AND AZINPHOS-METHYL IN WASHINGTON STATE

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ABSTRACT

The effects of field-aged residues of the new semi-synthetic spinosyn insecticide spinetoram (Dow AgroSciences, Indianapolis, IN) and the spinosad bait GF-120 (Dow AgroSciences, Indianapolis, IN) on mortality of apple maggot fly, *Rhagoletis pomonella* (Walsh), were determined in Washington State. Mortality caused by spinetoram (100 g a.i./ha) sprayed on apples and aged 7 d was significantly lower than that caused by fresh spinetoram. Spinetoram (100 and 75 g a.i./ha) aged for 7 d caused less than or as much mortality as spinosad (100 g a.i./ha) aged for 7 d. Fresh spinetoram and azinphos-methyl aged 7 or 14 d caused similar mortality, but aged spinetoram and spinosad caused lower mortality than azinphos-methyl. Apples treated with spinetoram (75 g a.i./ha) and with spinosad aged 7 d and exposed to flies produced a few larvae. However, even though spinetoram (100 g a.i./ha) aged 14 d did not kill all female flies, no larvae emerged from apples sprayed with this treatment. In separate tests, 0-d, 3-d, and 7-d old GF-120 on apple leaves caused greater mortality than 14-d old GF-120. Results show that spinetoram and GF-120 when fresh are highly toxic to *R. pomonella*, but that both have relatively short residual activities under the hot, dry conditions typical of central Washington in summer. Ingredients that prolong their toxicities or make their toxins available to flies longer may be needed to optimize their performance. Also, results suggest that adult fly mortality caused by spinetoram is not an accurate predictor of larval emergence from apples, and that possible non-lethal effects caused by spinetoram need to be examined.

Key Words: apple maggot, bait sprays, spinosyn formulations, organophosphate, residual activity

RESUMEN

Se determinaron los efectos de residuos envejecidos en el campo del insecticida spinosyn semi-sintético spinetoram (Dow AgroSciences, Indianapolis, IN) y el cebo de spinosad GF-120 (Dow AgroSciences, Indianapolis, IN), sobre la mortalidad de la mosca de la manzana, *Rhagoletis pomonella* (Walsh) en el estado de Washington (EEUU). La mortalidad causada por spinotoram (100 g i.a./ha) aplicada en manzanas y madurado por 7 dias fue significativamente más baja que la mortalidad causada por spinetoram fresco. Spinetoram (100 y 75 g i.a./ha) madurado por 7 d causó una mortalidad menor o igual que spinosad (100 g i.a../ha) madurado por 7 d. Spinetoram fresco y azinofosmetíl madurado por 7 o 14 d causó una mortalidad similar, pero el spinetoram madurado y spinosad causaron una mortalidad mas baja que azinofosmetíl. Las manzanas tratadas con spinetoram (75 g i.a./ha) y con spinosad madurado por 7 d y expuestas a moscas producieron muy pocas larvas. Sin embargo, aunque el spinetoram (100 g i.a./ha) madurado por 14 d no mató todas las hembras de moscas, ninguna larva emergió de manzanas rociadas con este tratamiento. En pruebas separadas de GF-120 madurado por 0-d, 3-d y 7-d, en hojas de manzana causó una mayor mortalidad que en GF-120 madurado por 14-d. Los resultados muestran que el spinetoram y GF-120 cuando estén frescos son altamente tóxicos al *R. pomonella*, pero ambos tienen una actividad residual relativamente corta bajo condiciones de alta temperatura y secas típicas del centro del estado de Washington en el verano. Ingredientes que prolongan su toxicidad o hagan que sus toxinas sean disponibles a las moscas por mas tiempo puede ser requeridos para optimizar su desempeño. También, los resultados sugerieron que la mortalidad de las moscas adultas causada por spinetoram no es un pronosticador preciso para la emergencia de larvas de las manzanas, y por otro lado se necesita examinar los posibles efectos no letales causados por spinetoram.

The apple maggot fly, *Rhagoletis pomonella* (Walsh), is a major pest of apple, *Malus domestica* (Borkh.) Borkh., in eastern North America and is an emerging pest in residential apple and hawthorn trees in central Washington State. The fly has established in low numbers in this region,

based on larvae found in hawthorn and apple on residential trees in 2003 and 2004. These findings resulted in export quarantines in areas within Kittitas and Yakima counties (Washington State Department of Agriculture 2005), which are part of the major apple-growing region in central Washington. The establishment of the fly in this region has major economic implications and threatens the export of commercial apples from Washington. The apple industry in Washington was estimated at US$1.11 billion in 2004 (Garibay 2005). There is a zero tolerance for *R. pomonella* larvae in apples transported within the state and to many overseas markets (Washington State Department of Agriculture 2001). Trap captures of *R. pomonella* adults in central Washington result in the spraying of host trees to suppress fly populations and reduce chances that flies will move into commercial apple orchards. Commercial apple orchards in central Washington to date have been free of *R. pomonella*.

Conventional insecticides are considered the leading candidates for controlling *R. pomonella* in central Washington. Currently, the organophosphate imidan (phosmet) is being used in residential trees in this region. Similarly, the organophosphates malathion and azinphos-methyl have been used for many years to control *R. pomonella* in the eastern (Neilson & Maxwell 1964; Neilson & Sanford 1974) and western U.S. (Mohammad & AliNiazee 1989). Despite the effectiveness of organophosphates, alternative materials are increasingly important because of the impending phaseout of organophosphate use due to the federal Food Quality & Protection Act (FQPA) (1996). Because of their relatively high mammalian toxicity, organophosphate insecticides may be hazardous to use, especially around homes or near water where fly-infested apple or hawthorn trees can occur. Effective organophosphates for controlling *R. pomonella* are not available for residential use.

Newer and safer insecticides need to be tested against *R. pomonella*. Laboratory bioassays with the newer insecticides imidacloprid, indoxacarb, pyriproxyfen, spinosad (85% spinosyn A and 15% spinosyn D), thiacloprid, and thiamethoxam showed that imidacloprid reduced oviposition the most and that imidacloprid and spinosad were the most toxic (Reissig 2003). However, these materials were not aged, and none of them appears to equal the organophosphates in toxicity. The residual toxicity after field aging for any material needs to be studied for several reasons. Some materials may be highly toxic initially when fresh but lose that toxicity quickly. This affects the frequency of insecticide applications and spray costs. No study has determined the effects of aging newer materials under the hot, dry central Washington conditions during summer on the toxicity of these materials to *R. pomonella*. In addition, even though resistance has never been documented in *R. pomonella*, overuse of one material invites the potential for increased tolerance to insecticides among non-target pests of apple, such as leafrollers and codling moth. Finally, the negative effects of insecticides on beneficial insects, which probably are exacerbated by frequent insecticide use, are well documented (e.g., Williams et al. 2003).

In this study, the objectives were to determine the effects of aging new or newer spinosyn insecticides, one incorporated into a bait mix, on the mortality of *R. pomonella*. Effects of aging insecticides on damage to apples caused by the flies were also determined. Tests focused on spinetoram, a new semi-synthetic spinosyn insecticide developed by Dow AgroSciences (Indianapolis, IN) that was accepted for expedited review under the United States Environmental Protection Agency's Reduced Risk Pesticide Program. Spinetoram is derived from fermentation products of the soil bacterium *Saccharopolyspora spinosa* Mertz and Yao, has a high safety profile, may have relatively long residual effects, and has never been tested against *R. pomonella*. Tests were also conducted with GF-120 Fruit Fly Bait (Dow AgroSciences, Indianapolis, IN), which contains spinosad, an insecticide that also has a high safety profile (Dow AgroSciences 2002). Results are discussed with respect to residual toxicities of these materials and their potential use in the management of *R. pomonella* in central Washington.

Materials and Methods

Effects of Field-Aging Spinetoram and Other Insecticides on Fly Mortality and Apple Fruit Damage

Experiment 1 compared a control and spinetoram and 2 other insecticides at various rates and ages: (1) untreated control, (2) a 100-g a.i./L suspension concentrate (SC) formulation of spinetoram at 172 mL/935 L water/ha (100 g a.i/ha), aged 0 d, (3) spinetoram at 100 g a.i./ha, aged 7 d, (4) spinetoram at 100 g a.i./ha, aged 14 d, (5) spinetoram at 127 mL/935 L water/ha (75 g a.i./ha), aged 7 d, (6) spinosad (Entrust® 80 WP, Dow AgroSciences, Indianapolis, IN) at 50.6 g/935 L water/ha (100 g a.i./ha), aged 7 d, (7) azinphos-methyl (Guthion® 50 WP, Gowan Company, Yuma, AZ) at 908 g/935 L water/ha (1,121 g a.i./ha), aged 7 d, and (8) azinphos-methyl at 1,121 g a.i./ha, aged 14 d. Spinosad and azinphos-methyl rates fell within recommended field rates. Materials were used within 1 year of receipt from the manufacturer.

'Fuji' apple trees at the United States Department of Agriculture, Agricultural Research Service (USDA, ARS) experimental orchard in Moxee, WA (46°33.23'N, 120°23.50'W) were sprayed with the various insecticide treatments in Jul and Aug 2004 at 7 or 14 d before exposure to flies. Control apples were from 1 tree and 0-d

treatment apples were from another tree. One treatment was sprayed on apples on each of 7 other trees with 1.18 liter RL Flo-Master® pressurized sprayers (Root-Lowell Manufacturing Co., Lowell, MI) until thorough coverage was achieved visually. Three different sets of trees were used or sprayed for 3 tests: test 1A, insecticides aged 13-27 Jul; test 1B, aged 3-17 Aug; and test 1C, aged 31 Aug to 14 Sep. Mean high and low temperatures and precipitation for tests 1A, 1B, and 1C were (1) 33.5°C and 12.4°C and 8 mm, (2) 31.5°C and 13.0°C and 9 mm (over 2 days), and (3) 25.7°C and 8.9°C and 2 mm (on 1 day), respectively. In tests 1A and 1B, rain occurred before 7-d sprays were made and in test 1C, it occurred after 7-d sprays were made. Most days were sunny, with the mean low humidity being ~30% and the daily mean being ~50%.

Three to 6 apples, each from a different branch, were removed from each tree ~5-6 h before testing. Apples were inserted through the calyxes into single upright nails on a board to keep them from rolling and contacting other surfaces and then transported to the Washington State University Research and Extension Center (WSUREC) in Puyallup in western Washington for the experiment. Quarantine restrictions precluded the maintenance of *R. pomonella* in central Washington for testing.

Flies were collected from feral and unmanaged apple trees in Puyallup in glass vials. Flies were maintained on dry 20% yeast extract (EZ Mix, Sigma, St. Louis, MO) and 80% sucrose (wt/wt) on paper strips ('food' hereafter) and on water in cotton wicks inside 3.8-liter cylindrical paper containers (17.5 cm high × 17.0 cm diameter) for up to 2 weeks before tests. This amount of time was needed to accumulate enough flies for tests. For testing, 1 control or treated apple was placed calyx end down on a shallow plastic dish inside a 3.8-liter container with food and water. For the 0-d treatment (fresh spinetoram at 100 g a.i./ha), apples were hung from a tree branch, sprayed, and dried for 1 h before being placed inside containers. Ten flies—6 males and 4 females in test 1A and 5 males and 5 females in tests 1B and 1C—were then transferred into a test container. Adult mortality was recorded from 1 to 10 d after exposure. Flies were recorded as dead if they could not walk when prodded or within 30 s of observation. Water vials were refilled every 2 d. Apple damage was measured as numbers of stings on fruit (from probing or oviposition) and larval emergence from fruit (an indication of larval infestation). Numbers of stings were counted under a microscope and apples were weighed and their circumferences measured at d 10. Each apple was then placed in a clear plastic 550-ml capacity container covered with organdy cloth. Numbers of larvae that emerged from the apples over 4 weeks were recorded. There were 3 replicates in test 1A, 6 in test 1B, and 3 in test 1C. Laboratory test conditions were 20-27°C and 40-50% RH under a 16 h L: 8 h D cycle.

Effects of Field-Aging GF-120 on Fly Mortality

Experiment 2 tested GF-120 Fruit Fly Bait, which is composed of 0.02% spinosad (wt/vol) (Dow AgroSciences 2002) mixed in a bait of Solulys corn protein, sugar, ammonium acetate, propylene glycol, and other ingredients (Thomas & Mangan 2005). An (1) untreated control and four age treatments of 40% GF-120 (vol/vol) (recommended rate, Dow AgroSciences 2002) were compared: (2) 0-d old, (3) 3-d old, (4) 7-d old, and (5) 14-d old. Trees used were 'Fuji' apple trees at the USDA, ARS orchard in Moxee. One tree provided control leaves and another tree provided 0-d treatment leaves. One age treatment was sprayed on each of 3 other trees. Leaves on 60-90 cm lengths of 5 branches of each tree were sprayed with ~10 mL of GF-120 in Jul and Aug 2004 with RL Flo-Master® pressurized sprayers. Two different sets of trees were used or sprayed for 2 tests: test 2A used GF-120 aged from 12-26 Jul and test 2B used GF-120 aged from 2-16 Aug. Mean high and low temperatures and precipitation during the 14 d of aging for tests 2A and 2B were (1) 29.8°C and 14.1°C and 8 mm (1 d), and (2) 31.9°C and 13.1°C and 9 mm (over 2 d), respectively. Most days were sunny, and humidity values were similar to those during aging for experiment 1. Three or 5 leaves, each from a different branch, were removed from each tree and placed inside plastic bags ~5-6 h before testing at the WSUREC in Puyallup.

Flies were held and tested inside 473-ml paper cartons (7 cm high × 5 cm diameter) (Neptune Paper Products, Newark, NJ) covered with organdy cloth and provided with food and water in the laboratory. Flies used in test 2A were reared from hawthorn fruit collected in 2003. Pupae had been chilled for 6 months at 4°C and then transferred to 20-27°C for adult emergence. Adults were held for 2-4 weeks before testing. Flies used in test 2B were collected from apple trees in Puyallup and held for 2-3 weeks before testing. For testing, a single control or treated leaf was placed inside a carton. The 0-d old treatment was a leaf sprayed with 1 mL of GF-120 1 h before testing. Ten flies (5 of each sex) were then introduced. Mortality was recorded daily, except for d 5 and 6 for both tests (weekends), up until d 10. There were 3 replicates of the control and treatments in test 2A and 5 replicates of each in test 2B. Laboratory test conditions were 25-27°C and ~40-50% RH under a 16 h L: 8 h D cycle.

Data Analyses and Statistics

Repeated-measures analysis of variance (ANOVA) was conducted on mortality data. One-

way ANOVA also was conducted on mortality data within days and on apple injury data. Data from d 1-4, 7, and 10 and from d 1-4 and 7-10 in experiments 1 and 2, respectively, are presented and analyzed. Because there were few replicates in tests 1A and 1B and because weather conditions during insecticide aging in these tests were similar, data from these were pooled for analyses. Test 1C was kept separate because of weather differences. Tests 2A and 2B also were pooled because there were few replicates per test and because weather conditions during GF-120 aging were similar. Percentages were square-root and arcsine-transformed before analyses. Sting and larval counts + 1 were subjected to square-root transformation. Means in one-way ANOVA were separated by using the Tukey test (SAS Institute 2001). This conservative test was chosen because of the high numbers of pairwise comparisons made.

RESULTS

Effects of Field-Aging Spinetoram and Other Insecticides on Fly Mortality and Apple Fruit Damage

Repeated-measures ANOVA indicated there were treatment and day differences, but there were also significant treatment × day interactions (tests 1A and 1B, $F = 16.5$, $df = 7, 408$, $P < 0.0001$; test 1C, $F = 17.0$, $df = 7, 126$, $P < 0.0001$), indicating patterns of mortality among the treatments differed on various days, even though mortality increased over time across all treatments. In tests 1A and 1B (Table 1), one-way ANOVA of data within days showed mortality caused by spinetoram (100) aged 7 d was significantly lower than that caused by fresh spinetoram (100), up until d 7. There were no differences in mortalities caused by spinetoram (100) aged 7 or 14 d until d 4-10, when 7-d old residues caused greater mortality. Spinetoram (75) aged 7 d was similar to spinetoram (100) aged 7 d but caused greater mortality than spinetoram (100) aged 14 d at d 4-10. Spinosad (100) aged 7 d caused greater mortality than spinetoram (100 and 75) aged 7 d at d 2 and 3, but caused similar mortality on other days. Fresh spinetoram usually was not different from aged azinphos-methyl within days. Mortality in tests 1A and 1B and in test 1C differed on 2 of the 6 d, and test number × treatment interactions occurred on 3 of the days (two-way ANOVA, $P < 0.05$). In test 1C (Table 2), similar to tests 1A and 1B, mortality caused by spinetoram (100) aged 7 d was significantly lower than that caused by fresh spinetoram, up until d 4-10. Also similar, in test 1C there were significantly greater mortalities caused by spinetoram (100) aged 7 than 14 d, this time on all 6 d of exposure, and spinetoram (75) aged 7 d caused lower mortality than spinetoram (100) aged 7 d on all days except d 10. Similar to tests 1A and 1B, spinetoram (75) aged 7 d caused greater mortality than spinetoram (100) aged 14 d at d 3 and 10. Unlike in tests 1A and 1B, however, in test 1C, spinosad (100) aged 7 d caused lower mortality than spinetoram (100) aged 7 d at d 1 and not other days, and greater mortality than spinetoram (75) aged 7 d at d 3-7, but not on other days. Fresh spinetoram usually was not different from aged azinphos-methyl within days.

Apples were smaller in tests 1A and 1B than in test 1C, but similar differences were seen in each analysis with respect to numbers of stings in ap-

TABLE 1. TESTS 1A AND 1B: MEAN CUMULATIVE PERCENT MORTALITY ± SE OF *RHAGOLETIS POMONELLA* EXPOSED CONTINUOUSLY TO 0-, 7- OR 14-D OLD INSECTICIDES ON APPLE FRUIT AT 1-10 D AFTER EXPOSURE IN THE LABORATORY.

Insecticides field-aged 13-27 Jul and 3-7 Aug 2004

Treatment		D 1	D 2	D 3	D 4	D 7	D 10
Control		0.0 ± 0.0 c	0.0 ± 0.0 d	0.0 ± 0.0 d	0.0 ± 0.0 d	2.2 ± 2.2 d	6.7 ± 5.5 c
Spinetoram (100) 0 d		36.7 ± 7.8 b	96.7 ± 1.7 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a
Spinetoram (100) 7 d		6.7 ± 2.4 c	14.4 ± 3.4 c	36.7 ± 8.7 c	47.8 ± 10.8 c	73.3 ± 11.9 ab	85.6 ± 9.3 a
Spinetoram (100) 14 d		5.6 ± 2.4 c	5.6 ± 2.4 cd	7.8 ± 4.3 cd	10.0 ± 4.4 d	24.4 ± 5.8 c	36.7 ± 7.1 b
Spinetoram (75) 7 d		5.6 ± 1.8 c	14.4 ± 5.0 c	28.9 ± 9.2 c	42.2 ± 9.8 c	67.8 ± 10.9 b	82.2 ± 6.8 a
Spinosad (100) 7 d		11.1 ± 4.8 c	43.3 ± 8.7 b	67.8 ± 11.5 b	72.2 ± 11.5 bc	91.1 ± 7.7 ab	95.6 ± 4.4 a
Azinphos-methyl (1,121) 7 d		76.7 ± 2.4 a	92.2 ± 1.5 a	93.3 ± 1.7 ab	95.6 ± 1.8 ab	98.9 ± 1.1 a	100.0 ± 0.0 a
Azinphos-methyl (1,121) 14 d		73.3 ± 3.7 a	94.4 ± 2.4a	95.6 ± 2.4a	98.9 ± 1.1a	100.0 ± 0.0 a	100.0 ± 0.0 a
One-Way ANOVA	*F*	47.1	77.6	41.1	39.0	34.1	44.9
df = 7, 64	*P*	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Tests pooled, 9 replicates of 10 flies each (test 1A: 6 males, 4 females, 3 replicates; test 1B: 5 of each sex, 6 replicates); 100 = 100 g a.i./ha; 75 = 75 g a.i./ha; 1,121 = 1,121 g a.i./ha.

Means followed by the same letter within days (columns) are not significantly different (Tukey test, $P > 0.05$).

TABLE 2. TEST 1C: MEAN CUMULATIVE PERCENT MORTALITY ± SE OF *RHAGOLETIS POMONELLA* EXPOSED CONTINUOUSLY TO 0-, 7- OR 14-D OLD INSECTICIDES ON APPLE FRUIT AT 1-10 D AFTER EXPOSURE IN THE LABORATORY.

Insecticides field-aged 31 Aug-14 Sep 2004

Treatment		D 1	D 2	D 3	D 4	D 7	D 10
Control		0.0 ± 0.0 c	0.0 ± 0.0 e	0.0 ± 0.0 e	0.0 ± 0.0 c	0.0 ± 0.0 c	3.3 ± 3.3 c
Spinetoram (100) 0 d		66.7 ± 8.8 a	96.7 ± 3.3 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a
Spinetoram (100) 7 d		20.0 ± 10.0 b	53.3 ± 8.8 bc	86.7 ± 3.3 b	90.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a
Spinetoram (100) 14 d		0.0 ± 0.0 c	0.0 ± 0.0 e	10.0 ± 0.0 d	33.3 ± 8.8 b	46.7 ± 13.3 b	53.3 ± 17.6 b
Spinetoram (75) 7 d		0.0 ± 0.0 c	16.7 ± 12.0 de	33.3 ± 3.3 c	56.7 ± 12.0 b	73.3 ± 12.0 b	90.0 ± 10.0 a
Spinosad (100) 7 d		0.0 ± 0.0 c	43.3 ± 3.3 cd	90.0 ± 0.0 b	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a
Azinphos-methyl (1,121) 7 d		80.0 ± 10.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a
Azinphos-methyl (1,121) 14 d		56.7 ± 6.7 a	86.7 ± 3.3 ab	93.3 ± 3.3 b	93.3 ± 3.3 a	100.0 ± 0.0 a	100.0 ± 0.0 a
One-way ANOVA	*F*	45.1	49.5	190.0	70.6	69.7	27.3
df = 7, 16	*P*	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Three replicates of 10 flies each (5 of each sex); 100 = 100 g a.i./ha; 75 = 75 g a.i./ha; 1,121 = 1,121 g a.i./ha.
Means followed by the same letter within days (columns) are not significantly different (Tukey test, $P > 0.05$).

ples and numbers of larvae that emerged from apples (Tables 3 and 4). In tests 1A and 1B (Table 3), spinetoram and other insecticides significantly reduced numbers of stings and numbers of larvae per apple by 90 to 100%. No larvae emerged from the spinetoram (100) treatments, including the 14-d old treatment, even though it killed only 36.7% of flies by d 10, with some females alive at that time. One larva emerged from an apple treated with spinetoram (75) and a total of 3 larvae emerged from 2 apples treated with spinosad. In test 1C (Table 4), there was a tendency toward reduced numbers of stings in all treatments (although not significant, according to the Tukey test) and all treatments prevented larval emergence from apples, including again from the spinetoram (100) aged 14 d treatment, even though it killed only 53.3% of flies.

Effects of Field-Aging GF-120 on Fly Mortality

Mortalities caused by 0-7 d old GF-120 were high, especially as days after exposure to flies increased (Table 5). Repeated-measures ANOVA resulted in a significant treatment × day interaction ($F = 22.0$, $df = 4, 303$, $P < 0.0001$), indicating the

TABLE 3. TESTS 1A AND 1B: MEAN APPLE SIZE AND APPLE DAMAGE ± SE CAUSED BY *RHAGOLETIS POMONELLA* EXPOSED CONTINUOUSLY TO 0-, 7- OR 14-D OLD INSECTICIDES ON APPLE FRUIT IN THE LABORATORY.

Insecticides field-aged 13-27 Jul and 3-7 Aug 2004

		Apple size		Apple damage	
Treatment		Weight (g)	Circ. (cm)	No. stings/apple	No. larvae/apple
Control		70.6 ± 5.8	16.9 ± 0.4	18.3 ± 6.0 a	3.9 ± 2.4 a
Spinetoram (100) 0 d		78.4 ± 8.7	17.3 ± 0.7	0.3 ± 0.2 b	0.0 ± 0.0 b
Spinetoram (100) 7 d		74.2 ± 7.5	17.1 ± 0.6	1.0 ± 0.5 b	0.0 ± 0.0 b
Spinetoram (100) 14 d		77.8 ± 6.8	17.5 ± 0.6	1.9 ± 0.3 b	0.0 ± 0.0 b
Spinetoram (75) 7 d		61.6 ± 5.8	16.2 ± 0.5	0.4 ± 0.2 b	0.1 ± 0.1 b
Spinosad (100) 7 d		67.3 ± 5.0	16.8 ± 0.5	0.9 ± 0.7 b	0.3 ± 0.2 ab
Azinphos-methyl (1,121) 7 d		82.5 ± 4.7	17.9 ± 0.4	0.1 ± 0.1 b	0.0 ± 0.0 b
Azinphos-methyl (1,121) 14 d		72.4 ± 4.4	17.2 ± 0.3	0.0 ± 0.0 b	0.0 ± 0.0 b
One-way ANOVA	*F*	—	—	14.8	2.7
df = 7, 64	*P*	—	—	<0.0001	0.0159

Tests pooled, 9 replicates of 10 flies each (test 1A: 6 males, 4 females, 3 replicates; test 1B: 5 of each sex, 6 replicates); 100 = 100 g a.i./ha; 75 = 75 g a.i./ha; 1,121 = 1,121 g a.i./ha. Circ., circumference.
Means followed by the same letter within apple damage measures (columns) are not significantly different (Tukey test, $P > 0.05$).

TABLE 4. TEST 1C: MEAN APPLE SIZE AND APPLE DAMAGE ± SE CAUSED BY *RHAGOLETIS POMONELLA* EXPOSED CONTINUOUSLY TO 0-, 7- OR 14-D OLD INSECTICIDES ON APPLE FRUIT IN THE LABORATORY.

Insecticides field-aged 31 Aug to 14 Sep 2004

Treatment		Apple size		Apple damage	
		Weight (g)	Circ. (cm)	No. stings/apple	No. larvae/apple
Control		112.8 ± 5.8	20.4 ± 0.1	24.3 ± 19.8	8.3 ± 6.3 a
Spinetoram (100) 0 d		111.7 ± 3.0	19.9 ± 0.2	0.0 ± 0.0	0.0 ± 0.0 b
Spinetoram (100) 7 d		113.5 ± 9.1	20.0 ± 0.5	0.0 ± 0.0	0.0 ± 0.0 b
Spinetoram (100) 14 d		119.1 ± 7.0	20.4 ± 0.4	0.0 ± 0.0	0.0 ± 0.0 b
Spinetoram (75) 7 d		111.5 ± 4.7	19.8 ± 0.3	0.0 ± 0.0	0.0 ± 0.0 b
Spinosad (100) 7 d		101.5 ± 5.7	19.4 ± 0.5	1.0 ± 0.6	0.0 ± 0.0 b
Azinphos-methyl (1,121) 7 d		123.0 ± 3.2	20.5 ± 0.3	0.0 ± 0.0	0.0 ± 0.0 b
Azinphos-methyl (1,121) 14 d		127.3 ± 6.2	20.8 ± 0.3	0.0 ± 0.0	0.0 ± 0.0 b
One-Way ANOVA	*F*	—	—	2.8	3.0
df = 7, 16	*P*	—	—	0.0412	0.0312

Three replicates of 10 flies each (5 of each sex); 100 = 100 g a.i./ha; 75 = 75 g a.i./ha; 1,121 = 1,121 g a.i./ha. Circ., circumference. Means followed by the same letter within apple damage measures (columns) are not significantly different (Tukey test, $P > 0.05$).

pattern of mortality among treatments differed at various days after exposure. There was almost no control mortality, large increases in mortality in 0-7 d old treatments, and small increases in mortality in the 14-d old treatment over time. One-way ANOVA indicated that the 14-d old GF-120 was not any more effective than the control until d 7 after exposure, and was never as effective as the 0-3 d old GF-120 during the 10 d (Table 5).

DISCUSSION

In experiment 1, we showed that the new insecticide spinetoram is highly toxic to *R. pomonella* when fresh, on a similar level to that of azinphos-methyl aged 7 to 14 d, but that its toxicity decreases rapidly after 7 d of aging in the field under the sunny, hot, and dry conditions typical of central Washington in Jul and Aug. By 14 d, spinetoram was no longer toxic to *R. pomonella*, except after 3 to 7 d of continuous exposure. Thus, spinetoram at 100 g a.i./ha would probably need to be applied at < 7-d intervals to be effective. The relatively short period of activity (compared with azinphos-methyl) suggests that spinetoram breaks down quickly in central Washington conditions and that ultraviolet light blocking or other agents need to be improved to prolong its toxicity. Also, it is possible that spinetoram was absorbed into plant tissue which occurs with spinosad (Dow

TABLE 5. TESTS 2A AND 2B: MEAN CUMULATIVE PERCENT MORTALITY ± SE OF *RHAGOLETIS POMONELLA* EXPOSED CONTINUOUSLY TO 0-, 3-, 7- OR 14-D OLD 40% GF-120 ON APPLE LEAVES AT 1-10 D AFTER EXPOSURE IN THE LABORATORY.

GF-120 Field-Aged 12-26 Jul and 2-16 Aug 2004

Days after Exposure	Age of GF-120 on apple leaves					One-way ANOVA	
	Control	0 D	3 D	7 D	14 D	*F* (*df* = 4, 35)	*P*
1	0.0 ± 0.0 b	47.5 ± 5.9 a	27.5 ± 4.9 a	25.0 ± 3.3 a	8.8 ± 4.0 b	23.3	<0.0001
2	0.0 ± 0.0 b	77.5 ± 8.2 a	55.0 ± 13.2 a	50.0 ± 9.6 a	12.5 ± 5.3 b	15.6	<0.0001
3	0.0 ± 0.0 b	86.3 ± 5.0 a	70.0 ± 9.1 a	75.0 ± 6.3 a	16.3 ± 6.3 b	30.6	<0.0001
4	0.0 ± 0.0 b	88.8 ± 4.8 a	78.8 ± 6.7 a	83.8 ± 5.6 a	17.5 ± 5.9 b	43.2	<0.0001
7	0.0 ± 0.0 c	100.0 ± 0.0 a	93.8 ± 3.2 a	96.3 ± 1.8 a	38.8 ± 12.0 b	73.1	<0.0001
8	0.0 ± 0.0 c	100.0 ± 0.0 a	98.8 ± 1.3 a	97.5 ± 1.6 a	42.5 ± 11.6 b	114.3	<0.0001
9	0.0 ± 0.0 c	100.0 ± 0.0 a	100.0 ± 0.0 a	97.5 ± 1.6 a	50.0 ± 14.4 b	68.3	<0.0001
10	1.3 ± 1.3 c	100.0 ± 0.0 a	100.0 ± 0.0 a	100.0 ± 0.0 a	53.8 ± 15.8 b	47.6	<0.0001

Tests pooled, 8 replicates of 10 flies each (5 of each sex).
Means followed by the same letter within days after exposure (rows) are not significantly different (Tukey test, $P > 0.05$).

AgroSciences 2004), making it unavailable to flies over time. If so, ingredients that prevent rapid absorption into leaves may help prolong its effectiveness. Spinosad in other studies shows decreased residual activity at 3-7 d after application (Williams et al. 2003), apparently similar to spinetoram. This suggests the different formulations of these 2 spinosyn insecticides do not affect the durations of their toxicity. Spinetoram used at rates described in this study might also be more effective if incorporated into a bait mix, similar to spinosad in GF-120, so that flies ingest more of it. Also, if a fly ingests degraded toxin, it may lead to quicker mortality than a fly that repeatedly contacts the degraded toxin.

The increases in mortality in spinetoram and other insecticide treatments over the 10 d of the tests suggest flies repeatedly contacted the apples as days progressed or that there was a delayed effect from 1 or a few initial contacts with the insecticide. Repeated contacts inside a cage may result in an overestimate of expected mortality under field conditions (Barry & Polavarapu 2005). Repeated contacts or a delayed effect may explain why fresh spinetoram was more toxic than spinetoram aged 7 d at d 1-4 and not 7 and 10 (tests 1A and 1B) or d 1-3 and not d 4-10 (test 1C).

Other results of tests 1A and 1B and of 1C were similar, with one key difference being the relative effectiveness of spinetoram versus spinosad. Overall results suggest spinetoram at 75 g a.i./ha is less effective than at 100 g a.i./ha, perhaps because the smaller amount is broken down more quickly than the larger amount, and that 75 g a.i./ha aged 7 d is more effective than 100 g a.i./ha aged 14 d, so aging may be more critical to effectiveness than the amount. In the one key difference, in tests 1A and 1B, spinosad at 100 g a.i./ha aged 7 d was more effective at d 2 and 3 than spinetoram at 100 g a.i./ha aged 7 d, but in test 1C, spinetoram was more effective than spinosad at d 1 and similar on all other days. The difference could be a result of warmer temperatures or ultraviolet light having a greater negative impact on spinetoram than spinosad over 7 d (5.8-7.8 ºC warmer in tests 1A and 1B than in test 1C). Another conclusion is that 100 g a.i./ha of spinosad, like this amount of spinetoram, is more effective than 75 a.i./ha spinetoram. Azinphos-methyl seemed less affected by warm temperatures or ultraviolet light than spinetoram and spinosad, based on its high effectiveness even after 14 d of aging.

The insecticides in this study did not kill *R. pomonella* adults quickly enough to prevent them from stinging apples. This included azinphos-methyl, albeit only one sting was detected in azinphos-methyl-treated fruit in all 3 tests combined. The inability of insecticides to prevent stinging or oviposition by *R. pomonella* agrees with earlier work with azinphos-methyl (Reissig et al. 1983) and indicates that insecticides must be applied before females are reproductively mature. Protection of apples from stings will therefore be problematic if treatment trees are surrounded by infested trees. However, overall results show that, despite the inability of spinetoram (100 g a.i./ha) to prevent stings, mortality and apple injury data did not lead to the same conclusions concerning effectiveness. Even though 14-d old spinetoram killed only 36.7 and 53.3% of flies after 10 d of exposure, the numbers of stings on the 14-d old spinetoram-treated apples were reduced 90 and 100% compared with controls, and no larvae emerged from any of these apples. This suggests there were non-lethal effects that reduced oviposition (some females were still alive at 10 d), including a repellent effect, or that flies stung the apples but did not oviposit. Eggs or larvae in fruit also may have been affected by spinetoram if it was absorbed into fruit tissue. Thus, mortality of adults is not the only variable to consider when evaluating the ability of spinetoram and the other insecticides to control *R. pomonella*. We did not cut apples and examine them for larvae. This could have resulted in the detection of internal damage in apples that had stings. It is possible some larvae died inside the fruit and did not emerge. If so, our data underestimated larval infestation rates.

In experiment 2 with GF-120, spinosad, similar to spinetoram, was highly toxic when fresh, but appeared to break down or was absorbed into leaves between 7-14 d under sunny, hot, and dry conditions. Whether bait components of GF-120 can be modified to protect spinosad against ultraviolet rays or to prevent rapid absorption into leaves under these conditions needs study. Results suggest that GF-120 in its present form needs to be applied every 7 d to be effective. Against the walnut husk fly, *Rhagoletis completa* Cresson, 20% GF-120 aged for only 3 d in hot weather in California lost toxicity (Van Steenwyk et al. 2003). The inconsistency of GF-120 in controlling *R. pomonella* in the eastern U.S.—ineffective in New York (Reissig 2003) and effective in 1 of 2 years in Michigan (Pelz et al. 2005)—may be caused in part by its short residual activity or by the high precipitation in the regions where it was tested. Residual toxicity of GF-120 in the eastern U.S. may more likely be reduced by rainfall than by dry conditions and high temperatures.

Our overall results indicate spinetoram and GF-120 when fresh are highly toxic to *R. pomonella* but that their residual toxicities need to be prolonged to optimize their performance against flies in central Washington. Direct comparisons of spinetoram and GF-120 are needed to determine whether one holds more promise than the other for fly control. Tests with both at higher rates, in different bait formulations, and in different spray volumes are needed to determine if they can be used in a management program for *R. pomonella*. Aged spinetoram is less toxic against adult flies than

azinphos-methyl, but it and GF-120 are more benign to the environment. Also, results suggest that adult fly mortality caused by spinetoram is not an accurate predictor of larval emergence from apples, and that possible non-lethal effects caused by spinetoram need to be examined.

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