Project Title: Fly Feeding Ecology and Food-Based Lures and Baits

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Budget History:

Item	Year 1: 2004	Year 2: 2005	Year 3: 2006
Salaries	18,000	18,000	18,000
Benefits	2,000	2,000	2,000
Wages	0	0	0
Benefits	0	0	0
Equipment	0	0	0
Supplies	2,000	87	87
Travel	0	0	0
Miscellaneous	0	0	0
Total	22,000	20,087	20,087

Objectives 2004-2006:
(1) Identify foods of western cherry fruit flies in nature.
(2) Determine when the flies feed, both daily and seasonally, and how much sugar and protein flies feed on in nature; amounts of foods in the environment.
(3) Determine the most attractive protein and sugar baits in the field and laboratory; baits that stimulate highest feeding and cause highest mortality.
4) Determine effects of enhancing baits with attractive compounds.
5) Determination of most effective bait sprays.

Significant Findings in 2004-2006:

● Feeding occurred mostly on leaf surfaces, with grazing behaviors most common. Bacteria and sugars on leaf surfaces are likely foods. Extrafloral leaf nectaries, cherry juice, and bird feces are food sources. The extensive grazing on leaf surfaces may result in flies finding baits frequently.

● Analyses of nutrients on leaf surfaces suggest flies need to forage over large areas to obtain their food requirements. This also suggests the extensive grazing behaviors result in chance encounters with baits and that this may be the main mechanism of fly control using baits.

● GF-20, Mazoferm, and Nulure protein and sugar baits tested were less attractive than ammonium hydroxide lures, suggesting attractiveness of baits need to be improved.

● Flies were not attracted to GF-120 or other baits from mid to long distances, suggesting mechanism of control is not through bringing flies in from around the tree.

● Flies were not attracted to GF-120, Mazoferm, or Nulure at close distances, suggesting the mechanism of control is not a close-range attraction.

● In the laboratory, mortality caused by GF-120, Mazoferm, Nulure, yeast (with Entrust [spinosad]), and Entrust alone was similar at all times post exposure to treatments, suggesting flies were encountering the drops through normal movement.

● Adding ammonia compounds to GF-120 increased its attraction in the field, suggesting attractiveness of baits can be enhanced.
● Adding ammonium acetate and ammonium carbonate did not increase feeding times by flies; feeding was longest on GF-120 alone or GF-120 with uric acid, suggesting some deterrence of feeding when ammonia compounds were added to baits.
● Mortality caused by GF-120 with or without enhancement with ammonia compounds was similar, suggesting that even short feeding times or contact with the baits (with spinosad) is sufficient to cause high mortality.
● GF-120, Mazoferm, and Nulure (with spinosad) sprayed on single cherry trees reduced the infestation levels of larval flies, but did not eliminate them; there was no evidence any one bait was superior to another. This suggests any bait with spinosad might have the same effect as GF-120.
● Spinosad alone sprayed on trees performed inconsistently; effective in one trial, not in the other. This suggests baits of any sort are more effective than Entrust alone, but this is unclear.

Methods 2004-2006:

(1) Identify Foods of Flies in Nature
In 2005, observations of feeding on different natural foods were made at one site in Zillah on 2 trees from 19 May to 12 June between 0830 and 1345 hours. In Roslyn, observations were made on 3 trees from 5 July to 4 August between 0900 and 1300 hours. A fly on a leaf or fruit was randomly selected and its feeding activities followed for a maximum of 10 min. The first fly that came into view and that could be watched from a distance of 15-25 cm was chosen. A timer was used to record the numbers and durations of fly feeding events on the leaves or fruit. The flies' mouthparts were observed closely. If a fly contacted the substrate with its proboscis, feeding was presumed to occur. Grazing consisted of rapid up and down movements of the mouthparts onto the substrate surface. Behaviors were recorded as: (1) grazing on undefined matter on leaves or fruit; (2) feeding on discrete substances: (a) nectar from extrafloral nectaries (EFNs), located on the distal part of the leaf petiole, 0-8 mm from the leaf; (b) cherry juice stains or splatters from damaged fruit; (c) bird feces, and (d) honeydew on or near aphid colonies. Attempts were made to follow at least 5 females and 5 males on leaves and fruit each sample day. A minimum observation of 30 sec was required for data to be included in analyses. At Zillah, flies were observed on 19, 24, 26, 27, and 31 May and 3, 6, 7, 10, 13, and 14 June. At Roslyn, flies were observed on 5, 7, 11, 14, 15, 18, 20, 25, 27, 29 July and 1, 3, and 4 August.

In 2006, methods for recording feeding on different substrates were similar to those in 2005, but there were also several differences: (1) to determine if flies grazed more frequently on top versus bottom of leaves, data of grazing on the 2 locations were kept separate; (2) to increase the numbers of flies observed, each fly was followed for a maximum of 5 instead of 10 min; (3) all flies were captured using a small glass vial after observations to reduce chances of repeated observations on the same fly; (4) observations were made earlier, usually between 0800-1100 hours, because 2005 observations suggested more flies foraged during this time than later. Flies were observed on 1, 7, 9, 12, 14, and 19 June. On 9 June, observations were made for one hour from 0600-0700 hours.

(2) Determine When Flies Feed, Daily and Seasonally
In 2005, 200 leaves and 200 cherries were collected from 3 trees on 23 May and 1, 8, 15, and 22 June. Leaves and fruit were dipped in water to remove sugars and other materials. Washings were placed inside bottles. At the same time, as many flies as possible were collected from the 3 trees using glass vials. Flies were immediately frozen in the field inside metal cans inserted in dry ice in Styrofoam boxes. All samples were then frozen at -80 ºC for later processing. Sugars and other substances from leaf and fruit washings and flies were analyzed using HPLC.

(3) Determine Most Attractive Baits; Baits that Stimulate Feeding and Cause Mortality
To test attraction of flies to ammonia and GF-120, Mazoferm, and Nulure, bait drops were applied on 5 leaves on the south sides of 3 to 6 cherry trees (0900-1500 hours) in Zillah and Roslyn in 2004.

The ammonia lure tested was a Nalgene bottle with a 0.05 cm hole. The bottle contained 10 ml of ammonium hydroxide saturated in cotton. Total bait volumes applied were 500 or 1,000 μl per 5 leaves. Flies seen feeding on the bait or within a 30 cm distance of the lure or baits were counted. For each test, fly numbers on leaves and fruit were recorded every 2 min for 30 min. After observations were made, leaves were removed and discarded. Branches were shaken to dislodge flies from the leaves or fruit. Positions of treatments were randomized after the first observation.

To determine long- to medium-range attraction to and feeding on baits, 2 tests were conducted in 2005 in Zillah using 40% concentrations of GF-120, Nulue, and Mazoferm. Test 1 was conducted using a total of 500 μl of water or baits: a volume of 100 μl water or bait (no spinosad) was applied on each of 5 randomly selected leaves. The control and each bait were applied on the south side of the same tree. Treatments were about 1-1.5 m apart, 1-2.5 m above the ground. Test 2 used 10 ml of the same solutions applied on a 45-60 cm stretch of a randomly selected branch with 20-40 leaves and fruit, using a 32-oz volume spray bottle. In both, numbers of female and male flies within 15 cm of droplets or that fed on the bait were recorded every 1.5-2 min for 30 min. Flies that clearly were the same at successive intervals were counted as one. All sprayed leaves were immediately removed after observations. Observations were made between 0900 to 1400 hours. An observer made observations of flies higher in trees while standing on a ladder. On each date, observations from one tree constituted a replicate, each with a control and one of each treatment. Test 1 was conducted on 24, 27, and 31 May. There were 5 replicate trees on 24 and 27 May and 3 trees on 31 May. Test 2 was conducted on 7, 10 and 14 June, with 3 replicate trees each day.

To determine close-range attraction, in 2005, one test was conducted in Zillah. The method used was to simulate situations where drops were detected by flies as they looked for food. About 25 μl of 40% GF-120, 40% Mazoferm, 40% Nulure or water were slowly placed 1-2 cm from a randomly selected fly on the top or underside of a leaf using a micropipette. This method was used in place of spraying because spraying would have resulted in direct contact of bait with the flies. Drops were placed in front of still or walking flies, but because flies sometimes moved, drops often ended up behind or on the side of flies. Most flies did not fly off or move when drops were placed near them, and thus did not appear to the observer to be disturbed by the presence of the observer and pipette tip. A fly was watched for a maximum of 5 minutes, and numbers and durations of all feeding events were recorded. Flies that flew off before 5 minutes were also used in analyses as long as they stayed a minimum of 15 seconds. After observations, flies were collected whenever possible using a glass vial to reduce chances that the same fly would be observed again. Treated leaves were removed. Observations were made 1-3 m above ground with the observer standing on a ladder if necessary. One to 3 trees were used on 25, 26, 27, 31 May and 2, 3, 6, 7, 9, 10, and 13 June. On each day, treatments were alternated so that each of the treatments and the control were tested before another set of the same materials were tested. There were 1 to 3 flies of each sex tested per treatment or control/day. Solutions were made fresh daily.

In 2006, the protocol to determine close-range attraction to baits was similar to that used in 2005, except for 3 differences. First, spinosad (Entrust) was added to all treatments. Second, in addition to a control and GF-120, Mazoferm, and Nulure treatments, spinosad alone was tested. Third, droplets were applied only to the top surface of leaves. Observations were made on 19, 22, 23, 24, 25, 26, 30, and 31 May and 5, 6, 8, and 9 June between 0800 and 1100 hours. There were large differences in weather during the roughly first half and second half of the season (the season based on 4 weeks when fly numbers are highest), unlike in 2005, so data from the 2 periods were compared.

4) Determine Effects of Enhancing Baits with Attractive Compounds

To test attraction of flies to ammonia and ammonia-enhanced GF-120, tests similar to those described for regular baits (above, objective 3) were done in Zillah and Roslyn in 2004. An ammonia lure was compared with Mazoferm + 10% ammonium carbonate (AC) (wt:wt), NuLure + 10% AC, and GF-120 + 10% AC.

In the laboratory, a test was conducted to determine the feeding responses of flies to enhanced bait. Water, GF-120, GF-120 + 10% uric acid (component of bird feces), GF-120 + 10% AA, GF-120 + 10% AC, and Entrust (spinosad) only were applied on an artificial leaf inside a half gallon cage. Five males and 5 females were released inside the cage. Observations were made over one hour of numbers of flies that fed, the feeding durations, and the time spent on the leaf not feeding. Another test was conducted to determine fly mortality caused by these treatments. A volume of 50 μl bait (as 3 drops) was placed on a dish on the bottom of a cage. A total of 30 flies was released inside cage. Mortality was determined at 2, 4, 6, 8, 24, and 48 hours after exposure.

5) Determination of Most Effective Bait Sprays

In 2005, 40% GF-120, 40% Mazoferm, and 40% Nulure were sprayed on residential cherry trees from May to June. Trees were 4-5 m tall and isolated or occurred in groups of 2 to 5. To determine the presence of and the approximate first emergence of flies, a sticky yellow panel was placed on each tree on 9 May and checked every day or 2 days for flies. Traps were baited with a lure containing 10 g of ammonium carbonate with two 1-mm holes. Three days after the first fly capture (on 13 May), the first application was made. Sprays were delivered using 1.18 liter RL Flo-Master© pressurized sprayers. For the GF-120 treatment, 90 ml of GF-120 was mixed in a total volume of 225 ml and applied on one tree (recommended rate for “spot spray of individual plants”). Mazoferm and Nulure were applied at the same rate. There were 8 control and 4 or 5 treatment trees. Sprays were applied as ~8 streaks using an upward motion around the periphery of each tree. Droplets varied in diameters, ~4-6 mm. Some of the “droplets’ were streaks of spray. Sprays were applied every 7 days, except once when it rained, in which case they were applied 3 days after a previous spray. Applications were made on 16, 19, 26 May, and 2, 9, and 16 June. Numbers of flies on traps were counted on all spray dates. Fruit from all trees were picked by 24 June. Fruit loads were low in most trees due to frost during fruit set in April and May, so a wide range in numbers of cherries were picked, from 14 to 506 per tree. For determining larval infestations, fruit were laid on hardware cloth on tubs and held outdoors. Numbers of pupae in the tubs after $\geq$ 30 days were recorded. Trees with traps that yielded no flies were dropped from the study.

In 2006, a spray protocol similar to that in 2005 was followed, except for the following, in order to match the feeding response test in 2006. First, in addition to a control and 40% GF-120, 40% Mazoferm, and 40% Nulure treatments, a spinosad (Entrust) only treatment was used, the same as in the feeding response test. Two tests were run. In test 1, a site in Zillah was used and 75 ml of spray was applied per tree, because trees were pruned and thus had much less foliage than trees used in test 2 (below). Each tree was 3-5 m tall. There were 3 replicate blocks of trees at this site, each with the control and 4 treatments. Blocks were different locations within the yard. In test 2, data from sites used in Zillah, Toppenish, and Yakima were pooled. The spray volume was 150 ml per tree. Each tree was 4-5.5 m tall. There were 7 control and 3 or 6 treatment trees. Only trees that had at least one fly captured on traps were used.

In both 2006 tests, the first sprays were made within 7 days of first fly capture. In test 1, traps were hung on trees on 8 May; there were 5 spray applications. In test 2, traps were hung 11 to 12 May; there were 5 to 7 applications. The range of applications was needed because there were different varieties of cherries with early or late developing fruit within the test. Also, birds threatened to remove all the cherries on some. Due to these factors, there was also variability in fruit picking dates, with 1 to 4 per tree. Unlike in 2005, trees bore heavy cherry fruit loads.

Results and Discussion

(1) Identify Foods of Flies in Nature

For simplicity, data over the season and not on a daily basis are shown. Grazing on leaves occurred much more frequently than feeding on cherry juice on leaves, bird feces on leaves, and extrafloral nectaries (EFNs) (Table 1). This was true on every date. Grazing occurred on every date, whereas feeding on EFNs, cherry juice, and bird feces was seen only on one, three, and one of the 11 dates, respectively. No aphid colonies were seen. On fruit, female and male flies rarely fed (Table 1). Males frequently stayed on fruit for entire 10-min observations. There were no differences between sexes (Table 1). In 2006, as in 2005, grazing on leaves occurred more frequently than feeding on cherry juice on leaves, bird feces on leaves, and EFNs (Table 1). This was true on every date. Grazing occurred on every date, but flies were seen feeding on EFNs, cherry juice, and bird feces only on 2, 4, and 3 of 6 dates, respectively. Nectar was seen in EFNs on every date. No aphid colonies were seen. The extensive grazing on leaf surfaces may result in flies finding baits.

Similarly, in Roslyn, of 49 females and 34 males on leaves, 10.2% and 11.8%, respectively, grazed leaves whereas none fed on EFNs and bird feces. Two females fed on cherry juice on leaves. Grazing and feeding on cherry juice were seen on 5 and 2 of the 13 dates, respectively.

Overall results suggest the extensive grazing behaviors observed can result in chance encounters with baits and that this may be the mechanism of control using baits.

(2) Determine When Flies Feed, Daily and Seasonally

Sugar analyses of flies throughout the season indicated consistently high levels, suggesting flies are able to find and feed on sugars regardless of the absence or presence of ripening cherries. Early analyses of sugars in the environment suggest diffuse food sources on cherry trees (flies and leaf samples have not all been processed and many are still frozen). The diffuse food sources may force flies to graze over large areas of the tree.

(3) Determine Most Attractive Baits; Baits that Stimulate Feeding and Cause Mortality

In the field, flies were not attracted to GF-120, Mazoferm, or Nulure baits from far distances, although they were to ammonium hydroxide lures (Table 2). A repeat of a similar test in 2005 revealed similar results: flies were not drawn to the GF-120 or Nulure and Mazoferm (Table 3). In 2005, when GF-120, Nulure, or Mazoferm were placed close to flies on leaves, flies were also not attracted to them (Table 4). In 2006, when spinosad (Entrust) was added, this same pattern was observed (Table 5). There was, however, a seasonal effect on fly responses. Feeding responses were greater during the second half of the season (Table 5). Results suggest that control should be similar using the different baits and that the baits should result in faster kill later in the season because either more flies respond to them or they respond more quickly to them. Results suggest there is no benefit of using GF-120 over Nulure or Mazoferm with spinosad.

In the laboratory, exposure of flies to GF-120, Nulure, Mazoferm, yeast, and Entrust all resulted in similarly high mortality of flies that were exposed to sugar only (Table 6). However, low mortality was seen across all treatments when flies were exposed to a sugar and yeast strip during the test (data not shown). The similar mortality among treatments is consistent with observations that none of the baits was superior to the others tested.

4) Determine Effects of Enhancing Baits with Attractive Compounds

When GF-120, Mazoferm, and Nulure were enhanced with ammonium carbonate, attraction to the baits was higher than to the control (Table 7). However, despite the greater attraction to enhanced GF-120, feeding on the GF-120 enhanced with AA or AC was not increased in the laboratory. In fact, the numbers of feeds and durations of feeds were highest on GF-120 alone and GF-120 + uric acid (Table 8). When flies were exposed to GF-120 alone or to GF-120 with ammonia compounds, mortality over time was similar (Table 9). The results suggest there are differences between attraction and feeding on ammonia-enhanced baits. An ideal enhanced bait should attract flies to the bait and once there, stimulate the flies to feed. However, it could be that even short feeding times (and therefore small amounts ingested) are sufficient to kill the flies.

5) Determination of Most Effective Bait Sprays

In 2005, when cherry trees were sprayed with 225 ml bait/tree, there were no differences in numbers of adult flies trapped among control and treatments, although numerically there were fewer flies in treatment than control trees (Table 4). Numbers of larvae per fruit were not significantly different in the control and the GF-120 treatment, but numbers in Nulure and Mazoferm treatments were significantly lower than in the control (Table 4). During the first 14 days of the test, there were 4 days of rain and 1.02 cm of precipitation. During the entire 40-day test (first spray to last fruit picking), there were 7 days of rain, for 1.25 cm total precipitation. Because there was relatively little rain, it is unlikely it affected results.

In 2006 in test 1, when trees were sprayed with 75 ml bait/tree, there were no significant differences in adult flies trapped and in larval infestations among control and treatment, although numerically there were fewer larvae per fruit in all treatments than in the control (Table 4). Larval infestations per fruit were much lower than in 2005. This was also true in test 2, when trees were sprayed with 150 ml bait/tree (Table 4). Fly populations in test 2 were lower than in test 1, and larval infestations were low even in control fruit. During the first 14 days of the test 1, there were 10 days of rain and 2.63 cm of precipitation. During the entire 36-day test (first spray to last fruit picking), there were 13 day of rain, for 3.17 cm total precipitation. For test 2, precipitation the first 14 days was the same as in test 1, but over the 50-day test, there were 15 days of rain, for a total of 3.32 cm precipitation. The rain may have affected results if they diluted the bait sprays.

The overall conclusion of the bait spray tests is that, under the fly densities and precipitation conditions encountered, the baits were unable to prevent larval infestation. At the very least, this could mean that the baits with spinosad did not kill the flies quickly enough to prevent egg laying. It is possible that after the flies laid the eggs, they fed on the spinosad in the baits and died. Whether the failure to prevent egg laying was the result of flies that matured (over 7 days) while on test trees and did not find the bait or the result of mature flies migrating in from surrounding trees was not determined. However, in isolated trees, the chances of this occurring seemed low.

Significance to the Industry and Potential Economic Benefits

The results of this project are significant to the cherry industry because they identify a mechanism of cherry fruit fly control using baits, explaining why GF-120, Mazoferm, and Nulure are similar in their effectiveness. Results using GF-120, Mazoferm, and NuLure show none is attractive and suggest flies find baits through normal foraging behavior rather than through a strong directed orientation towards odors. Identification of preferred foods for the flies is one step towards determining attractants or stimulants that can be incorporated into baits to make them more attractive and possibly more effective. Results suggest that use of Mazoferm and Nulure could reduce costs to growers, who need to spray baits often during the cherry season, especially when there is much rainfall. Additional work on the use of effective and long-lasting baits may help reduce spray frequencies and may further reduce chances larvae are ever found in fruit.

Table 1. Percentages of cherry fruit flies engaged in feeding on various substrates on sweet cherry leaves and fruit in over the season in 2005 and 2006, Zillah, WA

2005: On Leaves						
	Sex	N	Grazing On Leaf	EFN	Cherry Juice	Bird Feces
Season Totals	F	77	37.7	1.3	3.9	1.3
	M	70	22.9	1.4	4.3	0
F vs. M		X^2	3.76	-----	-----	-----
		P	0.0526	-----	-----	-----
2005: On Fruit						
	Sex	N	Grazing	Cherry Juice	Feces	
Season Totals	F	29	13.8	3.4	0	
	M	99	0	1.0	0	
2006: On Leaves						
	Sex	N	Grazing On Leaf	EFN	Cherry Juice	Bird Feces	EFN, Juice, Feces: X^2, *P*
Season Totals	F	131	45.0	1.5	6.9	6.1	4.57, 0.1016
	M	130	45.4	1.5	4.6	2.3	2.39, 0.3029
F vs. M		X^2	0.00	-----	0.60	2.27	
		P	1.0000	-----	0.4386	0.1317	

2005 - each fly observed for maximum of 10 min; 2006 – observed for 5 min; observations made between 0830 and 1430 hours (PST). EFN, extrafloral nectary; Data not analyzed when cells <5. 2005: season totals from 11 d. 2006: Season totals from 6 d.

Table 2. Effects of ammonium hydroxide lure and protein baits on mean numbers of cherry fruit flies attracted, May-June 2004, Zillah, WA

	Days[a]	Control	NH_3	Mazoferm	Nulure	GF-120
500 ul/leaf[b]	5	0.62	3.71	0.88	0.47	0.69
1,000 ul/leaf[c]	4	0.05	1.84	0.62	0.20	0.41

[a]3-6 replicate trees per day; [b]Zillah; [c]Roslyn

Table 3. Effects of baits applied on leaves of cherry trees on numbers of cherry fruit flies feeding on or near baits, May-June 2005, Zillah, WA

Test 1: 500 μl of bait spread on 5 leaves						
	No. Feeding			No. 15 cm From Bait, No Feeds		
Treatment	5/24	5/27	5/31	5/24	5/27	5/31
Water	0	0	0	0	0	0
40% GF-120	2	0	0	4	5	0
40% Nulure	0	0	0	2	0	4
40% Mazoferm	0	0	0	0	7	0
Test 2: 10 ml of bait sprayed on 20-40 leaves						
	No. Feeding			No. 15 cm From Bait, No Feeds		
Treatment	6/7	6/10	6/14	6/7	6/10	6/14
Water	0	0	0	4	5	1
40% GF-120	1	2	1	4	7	1
40% Nulure	1	0	1	5	0	1
40% Mazoferm	2	0	1	3	2	2

5 replicate trees on 24 and 27 May; 3 replicate trees on other dates.
On each date, recordings were made every 2 min for 30 min on each tree with the control and 3 treatments; totals of 39 females and 29 males recorded.

Table 4. Effects of placing bait droplets near cherry fruit flies on leaves on numbers of feeds and feed durations (min) on cherry trees, May-June 2005, Zillah, WA

Bait	Sex	*N*	% Response	No. Feeds	Feed Duration
Water Only	F	25	48.0	0.72	0.07
	M	20	35.0	0.80	0.10
Blank 40% GF-120	F	27	48.1	1.15	0.62
	M	20	50.0	1.05	0.50
Blank 40% Nulure	F	20	35.0	1.20	0.19
	M	21	42.9	0.33	0.09
Blank 40% Mazoferm	F	23	47.8	1.04	0.23
	M	20	25.0	0.30	0.03
2-Way ANOVA	Bait	df = 3, 168		$F = 0.93$; $P = 0.4252$	$F = 7.30$; $P = 0.0001$[a]
	Sex	df = 1, 168		$F = 1.57$; $P = 0.2124$	$F = 1.46$; $P = 0.2288$
	Bait × Sex	df = 3, 168		$F = 0.59$; $P = 0.6235$	$F = 0.37$; $P = 0.7770$

[a]Feeding duration on GF-120 > on water, Nulure, and Mazoferm.

Table 5. Effects of placing bait droplets near cherry fruit flies on leaves on numbers of feeds and feed durations (min) on cherry trees, May-June 2006, Zillah, WA

		N		% Response		No. Feeds	
Bait	Sex	Period 1	Period 2	Period 1	Period 2	Period 1	Period 2
Water Only	F	18	9	44.4	55.5	0.56	1.11
	M	17	24	47.1	62.5	0.71	1.04
40% GF-120	F	27	16	40.7	75.0	0.74	2.19
	M	12	19	16.7	57.9	0.33	1.26
40% Nulure	F	18	20	27.8	50.0	0.78	1.25
	M	23	17	21.8	58.8	0.43	1.24
40% Mazoferm	F	21	20	23.8	65.0	0.38	1.10
	M	16	15	18.8	66.7	0.19	1.00
Spinosad Only	F	14	16	7.1	68.8	0.07	1.38
	M	26	15	34.6	46.7	0.54	0.87
3-Way ANOVA	Bait		df = 4, 343			$F = 1.30$; $P = 0.2708$	
	Sex		df = 1, 343			$F = 1.72$; $P = 0.1905$	
	Period		df = 1, 343			$F = 39.59$; $P < 0.0001$	
	Bait × Sex		df = 4, 343			$F = 0.93$; $P = 0.4447$	
	Bait × Period		df = 4, 343			$F = 0.81$; $P = 0.5170$	
	Sex × Period		df = 1, 343			$F = 0.75$; $P = 0.3878$	
	Bait × Sex × Period		df = 4, 343			$F = 1.01$; $P = 0.4033$	

Each bait and the spinosad solution contained 0.0096% spinosad; Period 1, 19 to 30 May; Period 2, 31 May to 9 June.

Table 6. Cumulative percent mortality of cherry fruit flies exposed to various baits with spinosad at different times after exposure in the laboratory

Treatment	2 hours	4 hours	6 hours	8 hours	24 hours	48 hours
Water	0	0	0.7	0.8	1.6	2.7
GF-120	3.3	11.3	19.1	34.7	66.0	90.0
Nulure	2.7	8.7	29.3	46.7	88.0	97.3
Mazoferm	1.3	6.0	15.3	26.7	67.3	90.7
Yeast	0.7	10.7	22.7	42.7	78.0	94.7
Entrust	4.0	16.7	24.0	32.7	72.0	94.0
Blank GF-120 + Entrust	4.7	14.0	24.7	42.0	70.0	83.3

5 replicates, 30 flies each.

Table 7. Effects of adding ammonium carbonate (AC) on mean numbers of cherry fruit flies attracted to baits, Zillah, and Roslyn, 2004, WA

	Days[a]	Control	NH_3	Mazoferm +AC	Nulure +AC	GF-120 + AC
500 μl/leaf[b]	3	0.10	-----	0.98	1.33	0.32
1000 μl/leaf[c]	4	0.03	0.41	0.34	0.30	0.29

[a]Each day with 3 or 6 replicates; [b]Zillah; [c]Roslyn.

Table 8. Feeding responses of cherry fruit flies to GF-120 enhanced with various compounds in the laboratory

Treatment	No. Feeds	Feed Durations (min)	Dur. Non-Feeds (min)[a]
Control	0.17	0.01	2.55
GF-120	3.83	2.46	53.82
GF-120 + Uric Acid	4.17	2.06	28.30
GF-120 + AA	1.00	1.04	21.87
GF-120 + AC	0.33	0.07	20.93
Entrust Only	0.17	0.02	14.37

[a]On artificial leaf; 6 replicates each of 5 males and 5 females; AA, ammonium acetate; AC, ammonium carbonate.

Table 9. Cumulative mortality of cherry fruit flies exposed to GF-120 enhanced with various compounds at different times post exposure in the laboratory

Treatment	2 hours	4 hours	6 hours	8 hours	24 hours	48 hours
Control	0	0	0	0	3.3	20.0
GF-120	3.3	13.3	18.4	28.4	51.7	81.6
GF-120 + Uric Acid	1.7	10.0	20.8	34.2	51.6	76.6
GF-120 + AA	3.3	3.3	3.3	3.3	30.0	73.3
GF-120 + AC	2.5	5.0	7.5	20.0	51.7	76.7
Entrust Only	5.0	8.3	10.0	13.3	33.3	48.3

2-4 replicates per treatment; AA, ammonium acetate; AC, ammonium carbonate.

Table 10. Effects of bait sprays on mean numbers of cherry fruit fly adults and larvae from cherries using single tree replicates in Yakima County, WA, 2005 and 2006

2005: Yakima (16 May -24 June), 225 ml spray/tree					
Treatment	*N*	Flies/Trap[a]	Larvae/Fruit	% Fewer	No. Fruit Picked/Tree
Control	8	257.2	0.907a	-----	214.6
40% GF-120	4	16.0	0.501ab	45	73.8
40% Nulure	5	49.0	0.412b	55	233.4
40% Mazoferm	5	53.0	0.125b	86	273.2
1-way ANOVA df = 3, 18		$F = 2.31$ $P = 0.1111$	$F = 6.90$ $P = 0.0027$	-----	-----
2006: Test 1: Zillah (22 May – 26 June), 75 ml spray/tree					
Treatment	*N*	Flies/Trap[b]	Larvae/Fruit	% Fewer	No. Fruit Picked/Tree
Control	3	40.0	0.086	-----	194.3
40% GF-120	3	71.7	0.050	42	401.0
40% Nulure	3	115.7	0.004	95	496.7
40% Mazoferm	3	57.7	0.034	60	236.3
Spinosad Only	3	48.3	0.007	92	284.7
RBD ANOVA Df = 4, 8		$F = 1.22$ $P = 0.3760$	$F = 0.86$ $P = 0.5267$	-----	-----
2006 Test 2: Zillah, Toppenish, Yakima (22 May-10 July), 150 ml spray/tree					
Treatment	*N*	Flies/Trap[c]	Larvae/Fruit	% Fewer	No. Fruit Picked/Tree
Control	7	14.9	0.031	-----	368.0
40% GF-120	6	7.0	0.001	97	590.7
40% Nulure	6	6.0	0.004	87	576.0
40% Mazoferm	3	16.0	0.004	87	419.0
Spinosad Only	6	20.0	0.027	13	454.0
1-way ANOVA df = 4, 23		$F = 0.70$ $P = 0.6007$	$F = 2.04$ $P = 0.1215$	-----	-----

Each bait and spinosad solution contained 0.0096% spinosad.
Dates inside parentheses are first spray to last fruit picking; [a]over 42 d; [b]over 46 d; [c]over 57 d.
Means within columns followed by the same letter are not significantly different (LSD test, $P > 0.05$).