## A PEER REVIEWED PAPER

# **GREENHOUSE COMPARISON OF TRAPS AND LURES FOR FRUIT FLIES**

H. N. NIGG University of Florida, IFAS Citrus Research and Education Center 700 Experiment Station Road Lake Alfred, FL 33850-2299

S. E. SIMPSON Florida Department of Agriculture and Consumer Services Division of Plant Industry 3027 Lake Alfred Road Winter Haven, FL 33881

S. FRASER AND R. E. BURNS Florida Department of Agriculture and Consumer Services Caribbean Fruit Fly Mass Rearing Facility 1913 S.W. 34th Street Gainesville, FL 32608

Additional index words. Attractants, detection, eradication.

Abstract. A greenhouse testing system for fruit fly traps and lures was examined. The system allowed comparison of traps and lures for Anastrepha suspensa Loew within 14 days. Because the actual fly population is known each day of testing, the efficiency of the trap/lure can be estimated. The standard, glass McPhail trap baited with yeast pellets had an average daily efficiency of  $36.7 \pm 21.8\%$ . Greenhouse data for the standard, glass McPhail trap for female bias, lack of sugar attractiveness and general efficiency were comparable to previous field results. No trap/lure combination trapped all flies. The greenhouse fly population decreased about 50% per day regardless of trap numbers or lures. Costs for one greenhouse testing cycle were estimated as \$2,400 compared to \$52,500 for a similar test in the field.

The Caribbean fruit fly, Anastrepha suspensa Loew, has at least 84 host fruits in 23 plant families in Florida (Swanson and Baranowski, 1972). On June 4, 1974, the Japanese Ministry of Agriculture, Forestry and Fisheries found three pinhead-sized Diptera larvae in decaying white grapefruit from a 10,000 carton load. These were identified as an Anastrepha species (American Embassy, 1974; Nishimura, 1974; Rainwater, 1974). Consequently, Florida fruit for export to Japan were fumigated with ethylene dibromide (EDB). In 1983, the U. S. Environmental Protection Agency (EPA) canceled the use of EDB for domestic regulatory treatments of citrus (Simpson, 1993). In order to avoid the spiraling costs of fumigating fruit for export, the fly-free zone is an alternate, fumigation-free current practice (Simpson, 1993). The flyfree zone protocol requires, in part, removal of primary hosts and monitoring of A. suspensa with McPhail traps baited with a yeast/ borax/water combination.

*A. suspensa* attractant research efforts have been made with the male pheromone (Nation, 1972, 1975, 1983, 1989, 1990; Webb et al., 1983; Battiste et al., 1983; Robacker and Hart, 1987; Chuman et al., 1988), trap configurations including color (Perdomo et al., 1976; Burditt, 1982; Greany et al., 1982; Witherell, 1982; Davis et al., 1984; Sivinski, 1990; Barros et al., 1991), improved baits for traps (Sharp, 1987), and kairomones (Nigg et al., 1994).

Field testing of traps and lures is a time consuming, complicated and expensive process. Fruit fly populations must be located. The field numbers of flies and their sex ratio is never known. Sterile flies can be used in field tests. However, sterile flies may be too expensive for most researchers and may exhibit altered behavior. Once released in the field, the sex ratio and population of sterile flies changes daily. These numbers are very difficult, if not impossible, to determine in the field. Trap catches in the field may be low, making comparisons difficult, but in particular, the number of flies available to any individual trap is not known, making comparisons between individual traps invalid.

Field cages placed over host trees for fruit fly research have been used throughout the world by many researchers (Newman, 1928; Bateman and Morton, 1981; Morton and Bateman, 1981; Greany et al., 1982; Sivinski, 1990). With field cages experiments are limited, and need to be carefully planned and evaluated. The major limiting factors are weather, the availability of field sites, and the same difficulties found in a field test.

The purpose of this experiment was to evaluate a greenhouse testing system for comparing traps and lures for Tephritid fruit flies.

#### Materials and Methods

Insects. A. suspensa pupae were supplied by the Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville, Florida from the sterile fly rearing program. Fertile, laboratory-reared flies were shipped by overnight courier as 9-day-old pupae. These were held in an insectary at 24 to  $27^{\circ}$ C (75 to  $80^{\circ}$ F). Adults began emerging at 13 pupal days. Water as a 1% agar paddy and a yeast-sugar paddy on the top of each cage supplied water and food *ad libitum* (Nigg et al., 1995). Approximately 500 adults each were allowed to emerge in two cages (total ~1000 flies) (30.5 cm<sup>3</sup>, 12 × 12 inches, aluminum w/stocking front, BioQuip, Gardena, CA). At 4:30 PM on Sunday (day 0), these adult flies were released in a closed greenhouse without traps.

At 10:00 AM on Monday (day 1), the cages were closed and placed in a -17°C freezer for 4 hr. Empty pupal cases, unemerged flies, and dead adults in these cages were counted to provide the number of flies released. Fly age in each experiment ranged from 0-3 days on Monday and 4-7 days on Friday (day 5). In addition, about 200 adults each were allowed to emerge in the laboratory in three separate cages. These flies were killed in a freezer and counted to determine a sex ratio.

*Food experiment.* This experiment was conducted to estimate the survival time of flies in a greenhouse where neither supplemental food nor water was provided in most experiments. A five replicate experiment for different feeding regimes was conducted in plastic 32 oz. containers with a screened top. Each container was set up with 10 each newly emerged (about 4 hr old) males and fe-

Florida Agricultural Experiment Station Journal Series No. R-07008. This study was supported by a Florida citrus grower box tax for research. Funds for this project were made available from the Citrus Production Research Marketing Order by the Division of Marketing and Development, Florida Department of Agriculture and Consumer Services.

males. Treatments were no food and no water, water only, sugar only, yeast only, yeast and sugar, sugar and water, yeast and water, and yeast and sugar and water. Water was presented as a 15 gm, 1% agar patty. Agar patties were changed every day. Sugar was presented as a sugar cube. Brewer's yeast hydrolysate was prepared as a water yeast patty. This patty was microwaved until the water was driven out and the patty became a hard cake. The hard yeast cake was used as the yeast source in the yeast only treatment. The yeast and water and sugar treatment was presented as 1% agar patty, a sugar cube and a yeast-sugar-water patty. Dead flies were counted by sex every 24 hr. Treatments were monitored for 8 days.

Greenhouse description. The 7.1 m  $\times$  18.3 m (20'  $\times$  60', about 1/36th of an acre) glasshouse was temperature controlled at 75-80°F with two Arctic coolers and a gas fired heater. The house was sealed with duct tape for minor openings such as glass panels which had slid open and openings around plumbing pipes and the heater exhaust. The overhead vents were closed and sealed with weather stripping. The arctic coolers were not screened. However, these were inspected for flies in the funnel opening before, during and after each experiment and no flies were found. The entrance door remained closed during experiments except for the once a day fly counting and trap servicing. Sixty-nine containerized, 30 cm tall 'Valencia' citrus seedlings were placed in the house on plant benches prior to the first experiment and remained in the same position for all other experiments. Trees were watered at the base on Wednesday. Each tree was numbered. The leaves were removed from the first 15 cm (6") of the stem to aid in observing flies which usually rested on the leaf underside, as do other tephritid fruit flies (Malavasi et al., 1983). Three 3.8 cm diameter, 1.8 m (6') dowel rods were suspended from the ceiling, 2 m from the floor, parallel with the ends of the greenhouse with 3 m  $\times$  1.8 m (10'  $\times$  6') of black plastic shade cloth hung horizontally from the ceiling about 0.5 m (18") over each dowel to provide shaded areas. One dowel was suspended in the center of the greenhouse, and one dowel was suspended 3 m (10') from either end of the house. These dowels served as sites from which to hang traps and are referred to here as a trap location or blocks for statistical purposes. A glass thermometer suspended 1.4 m (4.5') off the floor and a recording hygrothermograph for temperature and humidity were placed in the middle of the house. On the first day after release (day 1) of an experiment and before traps were placed, flies were counted by plant number, north and south interior ceiling, heater bottom, dowel, and plastic shade cloth. Every part of the greenhouse was inspected for flies (e.g., undersides of benchtops, coolers, heater, resting on traps, etc.). On each experimental day, flies in the greenhouse were counted first. Counting generally took about 10 min with two observers and there was no fly movement which could interfere with this count. Then the traps were removed and fresh traps and lures were hung from the dowels. Trapped flies were counted and recorded by sex. These counts provided data for the distribution of flies, trap catch and the number of flies available for trapping. After the data were taken on day 5 (Friday), the remaining flies were killed by spraying all plant material and flies resting on other structures with 50% isopropanol. The greenhouse was ventilated until Sunday when the ridge vents were closed and the fly release process was repeated.

*Preliminary greenhouse experiments.* Three preliminary experiments were conducted with one yeast-baited McPhail trap baited with three, 5 g pellets in 250 ml of water placed on each bar before flies were released to determine if traps should be placed before or after flies were released. The 5 g yeast bait pellet consisted of four parts torula yeast and five parts dry technical grade (10 mole) borax decahydrate by weight. Flies immediately went to

traps placed before their release and did not distribute in the greenhouse. In order to allow for distribution of the population, flies were released on Sunday and traps were placed on Monday and subsequent days as described above.

*Experiments*. The traps and lures for each experiment are listed in Table 1. Experiments were run in the order listed in Table 1.

Experiment 1 compared plastic and glass McPhail traps. One trap with water and one trap with yeast pellets in 250 ml of water of each type were placed on each bar. Thus, each bar contained 4 traps; 12 traps total for the greenhouse. The glass McPhail can be either clear or light green glass. The plastic McPhail has an 'insect yellow' bottom. These traps previously were compared in the field by Barros et al. (1991). We wanted to see if our comparisons in the greenhouse were the same as Barros et al. (1991) and if McPhail traps caught more females in the greenhouse as they do in the field (Calkins et al., 1984; Epsky et al., 1993).

Experiments 2 and 3 examined the use of NuLure® in glass McPhail traps. Epsky et al. (1993) compared the attractiveness of NuLure® at different pH's in the laboratory and in the field. We wanted to determine if increasing the pH of NuLure® increased its attractiveness in the greenhouse. As we changed our traps and baits each day, decomposition of bait was not a factor as for Epsky et al. (1993). Experiment 2 compared glass and plastic McPhail traps as for experiment 1, except for the use of 250 ml of 10% NuLure®: 90% distilled water instead of yeast pellets and 250 ml of water. The pH of NuLure® was not altered for experiment 2. In experiment 3, NuLure® at four different pHs in glass McPhail traps was compared. Each bar contained 4 traps, 12 total for the greenhouse in experiments 2 and 3.

Experiment 4 compared the standard three yeast pellets and 250 ml of water to NuLure® at two pH's similar to Epsky et al.'s (1993) field comparison of NuLure® and yeast pellets. Each bar contained four traps including a water control.

Experiment 5 compared two Steiner traps baited with 100  $\mu$ l of  $\beta$ -pinene vs. one McPhail trap baited with yeast on each dowel, nine traps total for the house.  $\beta$ -pinene was a very attractive kairomone in our laboratory work with *A. suspensa* (Nigg et al., 1994). The Steiner trap is a 'dry' trap commonly baited with trimedlure for Mediterranean fruit fly (*Ceratitis capitata*, Wiedemann) (Steiner, 1957; Wong et al., 1982).

Experiment 6 compared yeast, yeast plus  $100 \ \mu l$  of  $\beta$ -pinene, 10% sucrose, and 10% sucrose plus  $100 \ \mu l$  of  $\beta$ -pinene to determine if  $\beta$ -pinene improved these baits. There were four traps on each bar.

Experiments 7, 8, and 12 were conducted to determine how the McPhail trap performed with no bait at all (water only). Experiment 7 had one McPhail trap with 250 ml of water per bar (3 traps total). Experiments 8 and 12 had four McPhail traps with 250 ml of water per bar (12 traps total).

Experiments 9, 10, and 11 assessed the performance of a single McPhail trap baited with yeast pellets in 250 ml of water.

Experiment 13 was conducted to determine if the color 'yellow' was attractive to *A. suspensa* in a Jackson trap configuration. The Jackson trap baited with trimedlure is the standard trap used for monitoring Mediterranean fruit fly.

Experiments 14 and 15 tested the attractiveness of NuLure® and the NuLure®-malathion (bait which is 20% malathion) used in tephritid fruit fly eradication programs and in the Florida fly free zone program (Simpson, 1993). For experiment 14, 9 cylindrical 0.3 m (12")  $\times$  1 m (39") plastic (acetate) traps were constructed. Each trap had an inward cone on each end of the cylinder with a 10 cm (4") opening on the inner end. Three traps were placed on each bar. Twenty milliliters of the NuLure® malathion bait combination were placed in a glass 9 cm petri dish in the center of each trap.

Table 1. Trap and lure comparisons in experiments in a greenhouse into which Caribbean fruit fly adults had been released.

Expt. no.	Trap type, number and lure
1	Plastic McPhail, 3 water (250 ml) and 3 yeast/borax pellets plus 250 ml water;* Glass McPhail 3 water and 3 yeast/borax pellets plus 250 ml water
2	Plastic McPhail, 3 water and 3 Nu-Lure® (10%, v/v), pH 4.46; Glass McPhail; 3 water and 3 Nu-Lure® (10%, v/v), pH 4.46
3	Glass McPhail; Nu-Lure® (10% v/v) 3 each, pH 4.46; 3, pH 7.02; 3, pH 8.02; and 3, pH 9.00 (adjusted with 4M NaOH)
4	Glass McPhail; 3 water (250 ml), 3 yeast/borax pellets plus 250 ml water; 1:10 Nu-Lure <sup>7</sup> :water 3 pH 4.47, and 3 pH 8.41 (adjusted with 0.1M 8.4 gly- cineglycine buffer)
5	Steiner traps; 6 baited with 100 ml of ß-pinene; Glass McPhail, 3 yeast/borax pellets plus 250 ml water
6	Glass McPhail; 3 yeast/borax pellets; 3 yeast/borax pellets + 100 µl β-pinene; three 10% sucrose + borax; three 10% sucrose + 100 µl β-pinene, all with 250 ml water
7	Glass McPhail; 3 water (250 ml)
8	Glass McPhail; 12 water (250 ml)
9	Glass McPhail; 1 yeast/borax pellets plus 250 ml water
10	Glass McPhail; 1 yeast/borax pellets plus 250 ml water
11	Glass McPhail; 1 yeast/borax pellets plus 250 ml water
12	Glass McPhail; 12 water (250 ml)
13	12 yellow, silver Jackson/tangle traps black
14	9 Malathion/NuLure® (20% Malathion); acetate traps
15	NuLure®; NuLure®/Malathion; NuLure®-20%/Malathion pH 7.14-3 days only
16	Glass McPhail; 1 yeast/borax pellets plus 250 ml water (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
17	Glass McPhail; 1 water (250 ml) (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
18	Glass McPhail; 1 yeast/borax pellets plus 250 ml water (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
19	Glass McPhail; 1 water (250 ml) (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
20	Glass McPhail; 1 yeast/borax pellets plus 250 ml water (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
21	Glass McPhail; 1 water (250 ml) (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)
22	Glass McPhail; 1 yeast/borax pellets plus 250 ml water (1 tree sprayed to runoff with 10% citrus molasses in each of the 3 zones)

\*Lopez et al. (1968, 1971), three, 5 g pellets in 250 ml water. The 5 g yeast bait pellet consisted of four parts torula yeast and five parts dry technical grade (10 mole) borax decahydrate by weight.

ter 3 days no flies had been caught so about 20,000 flies were released to see if any entered these traps. Experiment 15 was terminated after 3 days because of no fly catches.

Experiments 16 through 22 were similar to one another. Experiments 17, 19, and 21 assessed 1 McPhail trap with 250 ml of water suspended from the central dowel. Experiments 16, 18, 20, and 22 assessed 1 McPhail trap baited with yeast in 250 ml of water suspended from the central dowel. In each of these experiments, one tree in each of the tree zones was sprayed to runoff with citrus molasses (Florida Distillers, Lake Alfred, FL). The citrus molasses application was made to determine if a food source would affect the decline of the population, the trap catch of the yeast-baited McPhail traps or fly distribution.

Statistics and data analyses. For distribution analyses, the greenhouse was divided into three zones, east, middle, and west, with one zone per set of traps. Each zone contained 23 plants. The number of flies observed on plants was totaled for each zone. To test for equal distribution between zones and for differences in daily population declines, an ANOVA and Tukey's HSD Test (SAS Inst., Inc., 1989) were used. Trap catches and percent mortalities were compared by using GLM and Tukey's HSD Test (SAS Inst., Inc., 1989) with each trap location as a block and each trap as a treatment in a randomized block design. Percent dead data for males and females in the feeding experiment were compared within a day. Trap efficiencies were calculated by dividing the number of trapped flies by the number of flies in the greenhouse on the previous day times 100. The figure was produced with Sigma Plot (Jandel Scientific, 1992).

#### **Results and Discussion**

*Experiment 1.* There were no significant differences between total female or male flies trapped comparing glass and plastic McPhail traps (Table 2), nor were there differences when days

were compared (Table 3). Barros et al. (1991) found no difference in trap catches for *Anastrepha fraterculus* in the field comparing these same traps. Water-baited traps caught one or two flies per day in experiment 1. Our linear regression analyses confirmed that females captured in plastic and glass McPhail traps baited with yeast pellets plus 250 ml water were better predicators of observed populations than were captures of males (Table 2). These traps also caught more females than males (avg. 18 / vs. 4 ? per day, p =0.001) similar to field data on the use of McPhail traps (Calkins et al., 1984; Epsky et al., 1993).

Experiment 2, 3, and 4. Nu-Lure®, as presently formulated, was a very inefficient lure in McPhail traps, averaging about 2.4% of the observed population (Test No. 1, Table 4). Catches in Nu-Lure®-baited McPhail traps at pH 4.46, 7.02, 8.02, 9.00 in exper-

Table 2. Greenhouse Experiment 1: Linear regression analysis, observed and trapped by day for Caribbean fruit fly males, females, and total flies for plastic and glass McPhail traps baited with yeast/borax pellets in 250 ml of water.

Comparison	Formula	$\mathbb{R}^2$
1. Observed vs. Glass, Female	OBS = GT(6.52) + 35.30	0.9946*
2. Observed vs. Plastic, Total	OBS = PT(2.74) + 613.75	0.9870*
3. Observed vs. Glass, Total	OBS = GT(5.11) + 11.05	0.9469*
4. Observed vs. Plastic, Female	OBS = PT(2.92) + 79.06	0.9338*
5. Observed vs. Plastic, Male	OBS = PTM(11.41) + 30.81	0.4695
6. Observed vs. Glass, Male	OBS = GTM(7.96) + 72.12	0.2604

PTF = Plastic McPhail, total females, PTM = Plastic McPhail, total males, PT = Plastic McPhail, total flies, GTF = Glass McPhail, total females, GTM = Glass McPhail, total males, GT = Glass McPhail, total flies, OBS = observed.\*Statistically significant at a = 0.05, n = 3.

iment 3 were not different ( $\bar{x} = 8.7$  flies @ pH 4.46, 18.7 flies @ pH 7.02, 17.7 flies @ pH 8.02, 30.0 @ pH 9.00; df = 8,  $\alpha = 0.05$ ,

MSE = 37.1 Tukey's HSD test). In experiment 4, yeast averaged significantly more captured total, female, and male flies on day one than did Nu-Lure®, pH 4.47 and Nu-Lure® pH 8.41; total flies = 51 for yeast, 10 for NuLure® pH 8.41, 5 for Nu-Lure® pH 4.47; male flies = 17 for yeast, 3 for Nu-Lure® pH 8.41, 2 for Nu-Lure® pH 4.47; female flies = 34 for yeast, 7 for Nu-Lure® pH 8.41, 4 for Nu-Lure® pH 4.47. Yeast was different in each case at the 0.05 level by Tukey's HSD test. The fly capture of Nu-Lure® at different pHs were not different from one another. Average total flies per trap caught over the 4-day-period were 20 for yeast; 4 for Nu-

Table 3. Greenhouse Experiment 1: Correlation analysis of trapped and observed Caribbean fruit fly populations by sex in plastic and glass McPhail traps baited with yeast/borax pellets in 250 ml of water in a greenhouse.

Comparison	Pearson correlation coefficient
1. Observed vs. Plastic, total females.	0.99730*
2. Observed vs. Plastic, total males.	0.99349*
3. Observed vs. Plastic, total flies.	0.97307*
4. Observed vs. Glass, total females.	0.96632*
5. Observed vs. Glass, total males.	0.68521
6. Observed vs. Glass, total flies.	0.51025

\*Significant at  $\alpha = 0.05$ , n = 4 (days).

Lure® pH 4.47, 3 for Nu-Lure® pH 8.41, and 0 for water. These greenhouse data are comparable to the field comparison of yeast and Nu-Lure®-baited McPhail traps (Epsky et al., 1993).

*Experiments 5 and 6.* In experiment 5, the  $\beta$ -pinene baited Steiner traps caught no flies. In experiment 6 there were three 10% sucrose-baited McPhail traps and three 10% sucrose plus 100 µl  $\beta$ -pinene-baited McPhail traps. None of these traps caught flies. The unattractiveness of sugar as a bait agrees with Malavasi et al. (1990) with *A. grandis* (Macquart) and *A. fraterculus* (Wiedemann). There were no statistical differences between the trapping ability of yeast-borax and yeast-borax- $\beta$ -pinene baited traps in experiment 6 (yeast only total flies averaged 34 ± 22; yeast +  $\beta$ -pinene averaged 56 ± 10 means to S.D.; not different at  $\alpha = 0.05$ , Tukey's HSD test).

Table 4. Efficiency\* of McPhail traps in capturing Caribbean fruit fly adults 1 - 4 days following their release in a greenhouse.

Test no.	Lures and number of traps	Day	n	Single trap efficiency (Means ±SD)
1.	NuLure® (10%, 250 ml) 6 or12 McPhail traps	1	3	3.5 ± 2.0 a
		2	3	3.0 ± 1.3 a
		3	3	$2.2 \pm 1.6$ a
		4	3	$0.9\pm0.5~\mathrm{a}$
		Ove	rall	$2.4 \pm 1.5$
2.	Water (250 ml) 1-12 McPhail traps	1	6	$0.4 \pm 0.8  \mathrm{a}$
		2	6	$1.0 \pm 1.0 \ a$
		3	6	1.0 ± 1.3 a
		4	6	$2.9 \pm 2.5 a$
		Ove	rall	$1.3 \pm 1.5$
3.	Single glass McPhail with 3 Yeast/borax pellets in 250 ml water	1	7	41.2 ± 16.8 a
		2	7	37.4 ± 19.3 a
		3	7	34.1 ± 26.0 a
		4	7	34.0 ± 27.8 a
		Ove	rall	$36.7 \pm 21.8$
4.	Three glass McPail with 3 Yeast/borax pellets in 250 ml water	1	2	19.7 ± 5.1 a
	-	2	2	20.1 ± 4.5 a
		3	2	13.1 ± 8.1 a
		4	2	$14.2 \pm 0.8 \ a$
		Ove	rall	$16.8 \pm 5.4$
5.	Six glass McPhail with 3 Yeast/borax pellets in 250 ml water	1	2	8.0 ± 2.1 a
	-	2	2	11.7 ± 4.8 a
		3	2	$7.4 \pm 4.4$ a
		4	2	$5.0 \pm 0.2$ a
		Ove	rall	$8.0 \pm 3.4$

Means within each test followed by the same letter are not different at a = 0.05 by ANOVA and Tukey's HSD test. \*Efficiencies were calculated as the % captured of the observed population of the previous day's fly count, e.g., day 1 = Monday. Traps were replaced each succeeding day and trap efficiency calculated for the previous 24 hr.

Table 5. Cumulative percentage mortality of newly emerged Anastrepha suspensa adult flies on different feeding regimes.

	Day									
		1		2		3	2	1	4	4
Feeding regime	?	/	?	/	?	/	?	/	?	/
No food or water	$16 \pm 12 \text{ ab}$	$23 \pm 9 \ ab$	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a
Water	$6\pm 6$ b	$8\pm 8\ b$	$70\pm16~b$	$66\pm18~b$	$96 \pm 5 a$	94 ± 13 a	100 a	100 a	100 a	100 a
Sugar	$3 \pm 8 b$	$8\pm13$ b	$13 \pm 14 \text{ e}$	$39 \pm 24 \ cd$	$61 \pm 21 \text{ bc}$	$64 \pm 22 \ bc$	100 a	$98\pm 4\ a$	100 a	100 a
Yeast	$20\pm7~ab$	$40\pm10~a$	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a
Sugar + yeast	0 b	0 b	$16 \pm 13$ de	$10 \pm 7 e$	$62 \pm 11$ bc	$52 \pm 4 c$	$92 \pm 13$ a	$86 \pm 22$ a	100 a	100 a
Sugar + water	$4 \pm 6 b$	$24 \pm 33 \text{ ab}$	$51 \pm 14 \text{ bc}$	$66 \pm 5 b$	$82\pm20\ ab$	$97 \pm 7 a$	100 a	100 a	100 a	100 a
Yeast + water	$10\pm10\;b$	$10\pm14\ b$	$64 \pm 5 bc$	$70\pm0$ b	$96\pm5~a$	$98 \pm 4$ a	100 a	100 a	100 a	100 a
Yeast + sugar + water Yeast + sugar + water	$8 \pm 9 b$	8 ± 4 b	$10 \pm 12 \text{ e}$	$14 \pm 6 \text{ de}$	$10 \pm 12 \text{ d}$	$16 \pm 6 d$	$10 \pm 12 \text{ b}$	$18 \pm 5 b$	$\begin{array}{c} 13 \pm 13 \ b \\ 15 \pm 16 \ (day \ 8) \end{array}$	$19 \pm 6 b$ 21 ± 9 (day 8)

Mean  $\pm$  SD, n = 5, males and female means by day followed by the same letter are not different by GLM and Tukey's HSD test,  $\alpha = 0.5$ .

*Experiments 14 and 15.* No flies were captured in these malathion/Nu-Lure® (20% malathion) baited-traps even when thousands of adult flies were released into the greenhouse.

*Experiments 7 to 12 and 16 to 22.* Experiments 7, 8, and 12 contained 3 or 12 McPhail traps with only 250 ml of water. Experiments 17, 19, and 21 contained one McPhail trap with 250 ml of water. A few flies were trapped each day in these six experiments, leading to a trap efficiency of about 1.3% (Table 4, Test No. 2). The single yeast-baited trap in experiments 9, 10, 11, 16, 18, 20, and 22 had an efficiency of about 37% (Table 4, Test No. 3).

Feeding experiment. In this experiment, 100% of females and of males with no water and no food were dead within 48 hr (Table 5). Flies with yeast only were dead in 48 hr. Only flies provided with yeast, sugar, and water survived beyond 4 days (Table 5). Sugar only allowed survival until day 4 and was no different than sugar + yeast and sugar + water and was better than water, no food, no water and yeast alone and generally was better than yeast + water (Table 5). Newly emerged Caribbean fruit flies prefer baits with sugar (Nigg et al., 1995) and midgut protease activity necessary for yeast digestion is low in young Caribbean fruit flies (Yang et al., 2000). Apparently sugar is necessary for survival of young Caribbean fruit flies. These data suggest sugar should be included in baits for this fly. Because flies released into the greenhouse lived over the 5 day greenhouse experiments, they must have obtained food and water. We commonly observed flies lapping on leaf surfaces which might provide water and food.

*Fly distribution.* The ANOVA showed that the difference between the number of flies by day was highly significant (p = 0.0001) for all experiments, that is, the fly population was different each day of each experiment (F = 15.86, df = 20, p = 0.0001). Comparing zones, the difference in fly numbers was not significant in any experiment (F = 0.59, df = 2, p = 0.5596). Based on this equal fly distribution, each trap had an equal number of chances to catch a fly on each day of each experiment. Spraying one tree with 10% citrus molasses in each zone in experiments 16 to 22 (Table 1) did not result in flies gathering on these trees. Flies continued to distribute themselves evenly.

*Population decline.* We compared the greenhouse population decline rate of experiments with water baited traps only and yeast-baited traps only. These rates did not differ statistically on any given day. We then compared the decline rates of all 22 experiments. There were no statistical differences in these decline rates and, consequently, all data were combined to produce one population decline model (Fig. 1). The fly population in the greenhouse de-

Proc. Fla. State Hort. Soc. 113: 2000.

clined by about 50% per day regardless of traps, baits, or as in experiments 16 to 22, the provision of food in the greenhouse (Fig. 1).

*Trap efficiency*. We calculated the daily single trap efficiency (% of available population trapped) for Nu-Lure®, water, and yeast/borax pellet baits (Table 4). The Nu-Lure® and water baited traps were very inefficient, 2.3% and 1.3%, respectively (Table 4, Test Nos. 1 & 2, respectively). A single McPhail trap baited with yeast had an efficiency of about 37% (Table 4, Test No. 3). For a six yeast-baited trap experiment the single trap efficiency was about 8% (Table 4, Test No. 5). For a three yeast-baited trap experiment the single trap efficiency was about 8% (Table 4, Test No. 5). For a three yeast-baited trap experiment the single trap efficiency was about 8% (Table 4, Test No. 5). For a three yeast-baited trap experiment the single trap efficiency was about 17% (Table 4, Test No. 4). We multiplied the three and six trap individual experiment efficiencies by three and six, and reanalyzed our data. There were no statistical differences among the efficiencies in the one, three or six trap experiments (df = 2; day 1 - F = 0.00, p = 0.9492; day 2 - F = 1.23 p = 0.3003; day 3 - F = 0.88, p = 0.3758; day 4 - F = 1.10, p = 0.3248; no differences at  $\alpha = 0.05$  by Tukey's HSD test). A single

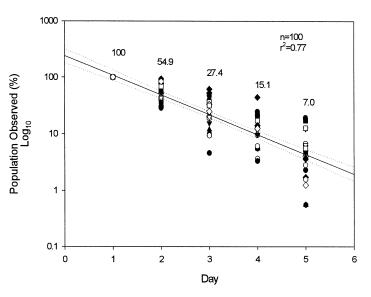


Figure 1. Percent of starting population (day 1) of Caribbean Fruit Flies remaining in the greenhouse each day, based on physical population counts each day before traps were removed and replaced. Counts began on Monday 16 hours after adult flies were released on Sunday. Symbols represent the 20 experiments whose data contribute to the decline curve.

McPhail trap apparently attracted flies from the entire greenhouse area and more traps split this catch (Table 4). This phenomenon also was seen in the field by Lopez et al. (1971) where yeast-baited McPhail traps at 25 traps per acre overlapped in their area of influence. This suggests that the efficiency of a trap could be estimated by placing one trap in the center of the greenhouse. However, the efficiency of a single yeast-baited McPhail trap catch averaged about 37% (see #3, Table 4). The daily population decline was 50%. On those days when a yeast-baited McPhail trap was present, an additional 13% of the flies disappeared, but not because they were trapped. Increased trap numbers did not change this relationship (Table 4, Fig. 1).

Our fly detection data disagree with the field data of Calkins et al. (1984). In their study the probability of detecting a fly with 1 trap per 0.4 ha and 900 released flies was 93%. In our experiments we released the equivalent of 1 million flies per ha which decreased to the equivalent of 1,000 flies per ha after 4 days. Our probability of detecting a fly was 100%. That is, on any day with a yeast-baited McPhail trap and with flies available, we captured a fly. When Calkins et. al (1984) released 9313 *A. suspensa* in 3.75 ha (about 2,500 flies/ha), about 13% were recovered with 168 traps. If we ratio our and the Calkins et al. (1984) field experiment

$$\left(\frac{1,000,000 \text{ flies/ha}}{2,5000 \text{ flies/ha}} \div 168 = 2.3 \times 13\% = 29.9\%\right)$$

we obtain a yeast-baited McPhail trap efficiency (equal basis) for the Calkins et al. (1984) field study of 29.9%. This is very comparable to our greenhouse efficiency of about 37%.

Our experiments showed that traps and lures for fruitflies can be compared in a greenhouse setting because flies distributed themselves evenly over pre-positioned resting areas (plants, ceiling, dowels, etc.), also a prerequisite for field experiments with released sterile flies (Calkins et al., 1984). Lures and traps previously tested in the field showed the same relative differences in the greenhouse (Calkins et al., 1984; Malavasi et al., 1990; Barros et al., 1991; Epsky et al., 1993). The yeast-baited McPhail trap had an overall efficiency of about 37%. A. suspensa populations declined in the greenhouse at about 50% per day regardless of trap numbers and lures. Other variables which can probably be evaluated in a greenhouse are: specific aged flies, lure age, fly behavior at bait stations, comparison of different species of flies, efficacy of pesticide/bait combinations, and bait station efficacy. Perhaps a greenhouse setting could not be used to test variable effects such as rainfall and wind on fly responses to traps and lures. We estimated the cost of testing one lure/trap combination in a greenhouse at \$2,400 over a period of 2 weeks (Table 6). The estimated cost of a similar test in the field is \$52,500. We estimate an approximate 10× savings in time, which is another advantage if traps and lures are tested in a greenhouse.

Our data suggest that greenhouse testing of fruit fly traps and lures before field testing is a valid screening technique which can provide comparisons of lures and traps more quickly, with less cost, and with more detailed data than field experiments.

### Table 6. Comparison of the estimated cost of field and greenhouse tests for fruit fly lures, traps, and insecticides

	Field*	Greenhouse**
Design and site selection:		
Personnel	\$ 2,400	N/A
Travel/vehicle	800	N/A
Pre application review/field set-up		
Personnel	1,000	N/A
Travel/vehicle	700	N/A
Application (×2 days)		
Personnel	4,000	\$ 100
Travel/vehicle	3,000	N/A
Aircraft	2,000	N/A
Flies/set-up/field release	1,700	200
rapping		
Personnel	1,200	300
Travel/vehicle	300	N/A
nalysis of Data		
Personnel	400	200
otal	\$17,500	\$ 800
epeated 3 times	\$52,500	\$2,400
cres/Facilities 4 reps $\times$ 10 acres $\times$ 6 treatments	240 Acres	1 greenhouse

\*6 Treatments, 4 replications each. Field estimated cost obtained from Don L. Harris, Chief, Bureau of Methods Development and Biological Control, FDACS—Division of Plant Industry, P.O. Box 147100, Gainesville, FL 32614-7100.

\*\*Cost estimated by authors.

#### Literature Cited

- American Embassy. 1974. Memorandum: Live larvae found in shipment U.S. grapefruit. Office of the Agricultural Attache, Foreign Agricultural Service, American Embassy, Tokyo, 5 June.
- Barros, M. D., P. M. do Amaral and A. Malavasi. 1991. Comparison of glass and plastic McPhail traps in the capture of the South American fruit fly, *Anastrepha fraterculus* (Diptera: Tephritidae) in Brazil. Fla. Entomol. 74:467-468.
- Bateman, M. A. and T. C. Morton. 1981. The importance of ammonia in proteinaceous attractants for fruit flies (Family: Tephritidae). Aust. J. Agr. Res. 32:883-903.
- Battiste, M. A., L. Strekowski, D. P. Vanderbilt, M. Visnick, R. W. King and J. L. Nation. 1983. Anastrephin and epianastrephin, novel lactone components isolated from the sex pheromone blend of male Caribbean and Mexican fruit flies. Tetrahedron Lett. 24:2611-2614.
- Burditt, A. K. 1982. Anastrepha suspensa (LOEW) (Diptera: Tephritidae) McPhail traps for survey and detection. Fla. Entomol. 65:367-373.
- Calkins, C. O., W. J. Schroeder and D. L. Chambers. 1984. Probability of detecting Caribbean fruit fly, Anastrepha suspensa (Loew) (Diptera: Tephritidae), populations with McPhail traps. J. Econ. Entomol. 77:198-201.
- Chuman, T., J. Sivinski, R. R. Heath, C. O. Calkins, J. H. Tumlinson, M. A. Battiste, R. L. Wydra, L. Strekowski and J. L. Nation. 1988. Suspensolide, a new macrolide component of male Caribbean fruit fly (*Anastrepha suspensa* [Loew]) volatiles. Tetrahedron Lett. 29:6561-6564.
- Davis, J. C., H. R. Agee and D. L. Chambers. 1984. Trap features that promote capture of the Caribbean fruit fly. J. Agr. Entomol. 1:236-248.
- Epsky, N. D., R. R. Heath, J. M. Sivinski, C.O. Calkins, R. M. Baranowski and A. H. Fritz. 1993. Evaluation of protein bait formulations for the Caribbean fruit fly (Diptera: Tephritidae) Fla. Entomol. 76:626-635.
- Greany, P. D., A. K. Burditt, Jr. and D. L. Chambers. 1982. Effectiveness of Jackson traps for fruit flies improved by addition of colored patterns. Scientific Notes. Fla. Entomol. 65:374-375.
- Jandel Scientific. 1992. Sigma Plot 5.0. Jandel Scientific 2591 Kerner Blvd., San Rafael, CA 94901.
- Lopez, F. D., L. M. Spishakoff and O. H. Becerril. 1968. Pelletized lures for trapping Mexican fruit fly. J. Econ. Entomol. 61:316-317.
- Lopez, F. D., L. F. Steins and F. R. Holbrook. 1971. A new yeast hydrolysate-borax bait for trapping Caribbean fruit fly. J. Econ. Entomol. 64:1541-1543.
- Malavasi, A., J. S. Morgantte and R. J. Prokopy. 1983. Distribution and activity of *Anastrepha fraterculus* (Diptera: Tephritidae) flies on host and non-host trees. Ann. Entomol. Soc. Amer. 76:286-292.
- Malavasi, A., A. L. Duate, G. Cabrini and M. Engelstein. 1990. Field evaluation of three baits for South American cucurbit fruit fly (Diptera: Tephritidae) using McPhail traps. Fla. Entomol. 73:510-512.
- Morton, T. C. and M. A. Bateman. 1981. Chemical studies on proteinaceous attractants for fruit flies, including the identification of volatile constituents. Aust. J. Agr. Res. 32:905-916.
- Nation, J. L. 1972. Courtship behavior and evidence for a sex attractant in the male Caribbean fruit fly, *Anastrepha suspensa*. Ann. Entomol. Soc. Amer. 65:1365-1367.
- Nation, J. L. 1975. The sex pheromone blend of Caribbean fruit fly males: isolation, biological activity, and partial chemical characterization. Environ. Entomol. 4:27-30.

- Nation, J. L. 1983. Sex pheromone of the Caribbean fruit fly: chemistry and field ecology. pp. 109-110. *In* IUPAC pesticide chemistry, human welfare and the environment, J. Miyamoto and P. C. Kearney (eds.), vol. 2 Pergamon, NY.
- Nation, J. L. 1989. The role of pheromones in the mating system of Anastrepha fruit flies. pp. 189-205. *In* Fruit flies: their biology, natural enemies and control, A. S. Robinson and G. Hooper (eds.). vol. 3A. Elsevier Science Publishers, Amsterdam.
- Nation, J. L. 1990. Biology of pheromone release by male Caribbean fruit flies, Anastrepha suspensa (Diptera: Tephritidae). J. Chem. Ecol. 16:553-572.
- Newman, L. J. 1928. Fruit-fly (Ceratitus capitata). Baiting and trapping experiments. West. Aust. Dep. Agr. Leaflet No. 244.
- Nigg, H. N., L. L. Mallory, S. E. Simpson, S. B. Callaham, J. P. Toth, S. Fraser, M. Klim, S. Nagy, J. L. Nation and J. A. Attaway. 1994. Caribbean Fruit Fly, *Anastrepha suspensa* (Loew) attraction to host fruit and host kairomones. J. Chem. Ecol. 20:727-743.
- Nigg, H. N., S. E. Simpson, J. A. Attaway, S. Fraser, E. Burns and R. C. Littell. 1995. Age-related Response of *Anastrepha suspensa* (Diptera: Tephritidae) to protein hydrolysate and sucrose. J. Econ. Entomol. 88:669-677.
- Nishimura, Y. 1974. Letter to D-N-E Sales, Inc., Ft. Pierce, FL, Quarantine rejection of grapefruits shipped per m/S "Arawak" due to presence of *fruit flies*. Sumitomo Shoji Kaisha Ltd., Tokyo, 7 June.
- Perdomo, A. J., J. L. Nation and R. M. Baranowski. 1976. Attraction of female and male Caribbean fruit flies to food-baited and male-baited traps under field conditions. Environ. Entomol. 5:1208-1210.
- Rainwater, H. I. 1974. Regulatory Services Office Memorandum: Florida exports to Japan. Pest Eradication and Control Administration, USDA, APHS, PPQ, 5 June.
- Robacker, D. C. and W. G. Hart. 1987. Electroantennograms of male and female Caribbean fruit flies (Diptera: Tephritidae) elicited by chemicals produced by males. Ann. Entomol. Soc. Amer. 80:508-512.
- SAS Inst., Inc. 1989. SAS/STAT User's guide, Version 6, 4th ed., Vol. 2, SAS Inst., Inc. Cary, NC. 846 pp.
- Sharp, J. L. 1987. Laboratory and field experiments to improve casein hydrolysate as an arrestant and attractant for Caribbean fruit fly, *Anastrepha suspensa* (Diptera: Tephritidae). Fla. Entomol. 70:225-233.
- Simpson, S. E. 1993. Caribbean fruit fly--free zone certification protocol in Florida (Diptera: Tephritidae). Fla. Entomol. 76:228-233.
- Sivinski, J. 1990. Colored spherical traps for capture of Caribbean fruit fly, Anastrepha suspensa. Fla. Entomol. 73:123-128.
- Steiner, L. F. 1957. Low-cost plastic fly trap. J. Econ. Entomol. 50:508-509.
- Swanson, S. W. and R. M. Baranowski. 1972. Host range and infestation by the Caribbean fruit fly, *Anastrepha suspensa* (Diptera: Tephritidae) in South Florida. Proc. Fla. State Hort. Soc. 85:271-274.
- Webb, J. C., T. Burk and J. Sivinski. 1983. Attraction of female Caribbean fruit flies, Anastrepha suspensa (Diptera: Tephritidae), to the presence of males and male-produced stimuli in field cages. Ann. Entomol. Soc. Amer. 76:996-998.
- Witherell, P. C. 1982. Efficacy of two types of survey traps for Caribbean fruit fly, Anastrepha suspensa (LOEW). Fla. Entomol. 65:580-581.
- Wong, T. T. Y., L. C. Whitehand, R. M. Kobayashi, K. Ohinata, N. Tanaka and E. J. Harris. 1982. Mediterranean Fruit Fly: Dispersal of Wild and Irradiated and Untreated Laboratory-Reared Males. Environ Entomol. 11:339-343.
- Yang, L. K., H. N. Nigg, S. Fraser, E. Burns and S. E. Simpson. 2000. Midget proteinase types and sodium tetraborate effects on midget proteinase activities of female *Anastrepha suspensa* (Diptera: Tephritidae). Ann. Entomol. Soc. Amer. 93:602-609.