

## CAPTURE OF *ANASTREPHA SUSPENS*A AND STERILE MALE *CERATITIS CAPITATA* (DIPTERA: TEPHRITIDAE) IN MULTILURE TRAPS VERSUS PHASE 4 TRAPS

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### ABSTRACT

Field trials were conducted in south Florida to compare capture of wild Caribbean fruit flies, *Anastrepha suspensa* (Loew), and sterile male Mediterranean fruit flies, *Ceratitidis capitata* (Wiedemann), in Multilure traps, which are McPhail-type traps that use an aqueous solution to retain attracted flies, and Phase 4 traps, which are open-bottom dry traps that use a sticky insert to retain attracted flies. One study was conducted in a guava orchard and compared capture of *A. suspensa* in both trap types baited with ammonium acetate plus putrescine alone (two-component BioLure) or in combination with trimethylamine (three-component BioLure). A second study compared captures of *A. suspensa* and sterile male *C. capitata* in traps baited with three-component BioLure in an urban area near the end of the eradication program for a *C. capitata* outbreak. In both studies, captures were higher in the Multilure traps than the Phase 4 traps baited with the same lure, with catches ranging from 5:1 for sterile *C. capitata*, and ~10:1 to ~100:1 for wild *A. suspensa*. Large scale area-wide deployment of fruit fly detection traps is costly in both materials and in the time and effort required in routine servicing. Although a simpler and cheaper trap such as the Phase 4 trap would be a welcome relief to any large scale area-wide detection programs, it must perform effectively. This is the first report of tests of fruit fly capture in Phase 4 traps conducted under of south Florida conditions.

Key Words: Mediterranean fruit fly, Caribbean fruit fly, detection, monitoring, trap

### RESUMEN

Se realizaron ensayos de campo en el sur de la Florida para comparar la captura de moscas salvajes de la mosca del Caribe de la fruta, *Anastrepha suspensa* (Loew), y machos estériles de la mosca mediterránea de la fruta, *Ceratitidis capitata* (Wiedemann), en trampas Multilure, que son trampas del tipo McPhail que utilizan una solución acuosa para retener moscas atraídas, y trampas de Fase 4, que son trampas secas de fondo abierto que utilizan una banda pegajosa para retener las moscas atraídas. Se realizó un estudio en un huerto de guayaba y se comparó la captura de *A. suspensa* en ambos tipos de trampas cebadas con acetato de amonio más solo putrescina (BioLure de dos componentes) o en combinación con trimethylamine (BioLure de tres componentes). Un segundo estudio comparó la captura de *A. suspensa* y machos estériles de *C. capitata* en trampas cebadas con BioLure de tres componentes en una zona urbana cerca del final del programa de erradicación de un brote de *C. capitata*. En ambos estudios, las trampas Multilure capturaron mas moscas que la trampas de Fase 4 cebadas con el mismo señuelo, y esto varió de 5:1 para la captura de machos estériles de *C. capitata* y de ~10:1 a 100:1 para las *A. suspensa* silvestres. La implementación de trampas de detección de moscas de la fruta en gran escala de área muy amplia es costosa, tanto en materiales como en el tiempo y el esfuerzo necesarios para el mantenimiento rutinario. Aunque una trampa

simple y barata como la trampa de Fase 4 sería un alivio para cualquier programa de detección en gran escala de toda la zona, esta debe ser realizada con eficacia. Este es el primer informe de las pruebas de la captura de moscas de la fruta en trampas de Fase 4 realizadas bajo las condiciones del sur de Florida.

Palabras Clave: mosca mediterránea de la fruta, mosca Caribe de la fruta, detección, monitoreo, trampa

Tephritid fruit flies are important target pests for trapping programs conducted by combined regulatory agencies in Florida under the Cooperative Fruit Fly Detection Program. The Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), is the primary target pest for these trapping activities due to its importance worldwide and its threat to US agriculture. The area-wide trapping program is also used to detect a number of *Bactrocera* and *Anastrepha* species fruit flies that threaten to invade Florida, specifically the oriental fruit fly, *Bactrocera dorsalis* (Hendel); the melon fruit fly, *Bactrocera cucurbitae* (Coquillett); the Mexican fruit fly, *Anastrepha ludens* (Loew); the West Indian fruit fly, *Anastrepha obliqua* (Macquart); the South American fruit fly, *Anastrepha fraterculus* (Wiedemann); and the guava fruit fly, *Anastrepha striata* (Schiner) (Anonymous 2002). An area of approximately 22,000 km<sup>2</sup> in Florida involving 43 counties that are historically at risk for fruit fly introductions are routinely surveyed with approximately 55,000 fruit fly detection traps year round. Multilure traps baited with three-component food-based BioLure (Suterra LLC, Bend, Oregon) (ammonium acetate, putrescine and trimethylamine) and Jackson traps baited with trimedlure are the standard traps used in Florida for detection of *C. capitata*. Multilure traps make up about 48% of the traps used in the preventive sterile *C. capitata* release program areas and 18% of the total fruit fly detection traps deployed in the state. Advantages of the Multilure plus BioLure traps are that they capture female *C. capitata*, which are rarely captured in trimedlure-baited traps (Nakagawa et al. 1970), and they have been found to detect new invasions of *C. capitata* before the trimedlure-baited traps (Papadopoulos et al. 2001). They also capture female *Bactrocera* and *Anastrepha* species fruit flies, although it has been shown that for *Anastrepha* species they may not be as effective as Multilure traps baited with either two-component BioLure (ammonium acetate and putrescine) or with liquid protein baits such as torula yeast/borax (Epsky et al. 2003; Holler et al. 2006; Epsky et al. 2011).

Fruit fly traps that use a liquid to retain captured flies are costly and time consuming to maintain in the field. A simple disposable female-targeted dry trap could be more cost effective and less effort to maintain at the numbers needed for area-wide detection. It was hoped that identification of synthetic food-based lures would lead to development of such a dry trap, and initial testing used

a closed-bottom dry trap with a toxicant square to retain attracted flies (Heath et al. 1995). Problems with handling these traps led to the development of an open-bottom dry trap that used a sticky panel to retain attracted flies (Heath et al. 1996). Success in using this trap in Guatemala, especially in the mountainous coffee growing regions that makes it difficult to deploy aqueous traps, led to production and use of a version of an open-bottom dry trap, called the Phase 4 trap (e.g., McQuate & Peck 2000; Villaseñor et al. 2000; Midgarden et al. 2004). Tests of *C. capitata* capture in traps baited with the three-component BioLure that were conducted in Spain and Turkey found that open-bottom dry traps were less effective than McPhail-type traps with aqueous solutions, although there was equal capture in tests conducted in Honduras (Epsky et al. 1999). A test of the open-bottom dry traps with *A. ludens* in Mexico also found that they were less effective (Thomas et al. 2010).

Few tests of fruit fly female-targeted dry traps have been conducted in Florida. The Caribbean fruit fly, *A. suspensa* (Loew), is established in Florida and is a target pest for trapping protocols under the Caribbean fruit fly-free zone certification protocol (Greany & Riherd 1993; Simpson 1993). Early tests of two-component BioLure in closed-bottom dry traps found low capture (N.D.E., unpublished data), although these same lures in Multilure traps are highly effective (Hall et al. 2005; Epsky et al. 2011). There have been no tests of the open-bottom dry trap for either *A. suspensa* or *C. capitata* for conditions in south Florida.

Therefore, 2 studies were conducted in south Florida to compare capture in Multilure traps versus Phase 4 traps. The first study compared capture of wild *A. suspensa* with traps baited with the *Anastrepha*-targeted two-component BioLure or the *C. capitata*-targeted three-component BioLure in tests conducted in a guava planting. The second study compared captures of sterile male *C. capitata* and wild *A. suspensa* in traps baited with three-component BioLure in an urban area near the end of the eradication program for a *C. capitata* outbreak.

## MATERIALS AND METHODS

### Traps and Lures

Traps used in this study included Multilure traps (Better World Manufacturing Inc., Fresno, Califor-

nia) and Phase 4 traps, which are open-bottom dry traps (Heath et al. 1996; IAEA 2003) that are produced for Moscamed in Guatemala for their *C. capitata* detection program. The synthetic attractants used were individual component BioLure formulations of ammonium acetate, putrescine, and trimethylamine (Suterra LLC, Bend, OR). Attractants tested were the combination of ammonium acetate and putrescine lures, which targets *A. suspensa* (Epsky et al. 2011), or the combination with ammonium acetate, putrescine, and trimethylamine, which targets *C. capitata* (Heath et al. 1997). Multilure traps contained 300 mL 10% polypropylene glycol (vol:vol; LowTox, Prestone, Danbury, Connecticut) aqueous solution to retain captured flies. Phase 4 traps (9 cm diam × 15 cm height) are made from an opaque green waxed cardboard with 3 holes (2 cm diam) evenly spaced around the midline of the trap. A yellow sticky panel (7.6 × 12.7 cm; Suterra LLC), coated on both sides, was suspended inside the trap to retain captured flies.

#### Comparison of Multilure versus Phase 4 Traps with Two- or Three-Component BioLure.

A field test was conducted in a guava orchard, *Psidium guajava* L., at the Univ. Florida, Tropical Research and Education Center (TREC) in Homestead, Florida. Guava was in fruit and *A. suspensa* adults were the only tephritids sampled in these tests. There were 4 treatments that included both trap types baited with both lure combinations. There was 1 trap per tree, with the traps placed in 5 rows (replicates) of trees, for a total of 20 traps sampled. There were at least 10 m between rows and 10 m between traps within a row, and the test was initiated 6 Oct 2010. Due to inclement weather, traps were initially sampled after 14 days, but then were sampled every 7 days for a total of 4 sampling periods over 5 wk. At the time of sampling, the sticky inserts were replaced, the retention fluid was recycled and additional liquid added to replace liquid lost to evaporation, and the numbers of males and females were recorded. The synthetic lures were not replaced throughout the study. A complete randomized design was used for initial trap placement, and traps were rotated sequentially to the next position within a block at time of sampling, so that all treatments were tested in all positions within a block. Numbers of female and male *A. suspensa* per trap were converted to flies per trap per day per block.

#### Comparison of Multilure versus Phase 4 Traps with Three-Component BioLure

At the recommendation of the 2010 Boca Raton Medfly Eradication Program Technical

Working Group, a test was conducted in Palm Beach County to evaluate the potential incorporation of the Phase 4 dry female trapping system into the Cooperative Fruit Fly Detection Program as a cost saving alternative. The test was initiated just prior to the officially declared eradication in a 2600 km<sup>2</sup> area under sterile Mediterranean fruit fly aerial release. One hundred Public Land Survey sections (each 2.6 km<sup>2</sup>) within this area were used as blocks, and a single Phase 4 trap was paired with the standard fruit fly detection Multilure traps in the same section. All traps were baited with three-component lure and were serviced weekly following standard fruit fly detection procedures specified in the Mediterranean Fruit Fly Action Plan for Florida 2002. Since this was a test to compare the effect of trap design on fruit fly captures, within a section the Multilure and Phase 4 traps were placed on opposite sides of the same host tree, and each section was used as a block for the analysis. Due to the size of the area, the number of agencies and personnel involved, and the trapping transitions occurring during the termination of the eradication program, the number of weekly traps services varied between 3-4 wk. Numbers of sterile male *C. capitata*, and wild female and male *A. suspensa* per trap were counted weekly and converted to flies per trap per day per block.

The ability to detect the presence of target economic fruit flies with a high level of certainty is of great importance to any fruit fly detection program. Fruit fly detection traps and survey methods need to be evaluated for their effectiveness in detecting the presence of at least one individual per sampling unit. Therefore, a method using maximum likelihood for estimates of occupancy and detection probabilities employing site survey detection history was employed using the computer software program PRESENCE (MacKenzie et al. 2002; USGS 2002). Traditional presence-absence methods based on point data are known to underestimate actual presence because of "false absences" due lack of detection when the species is actually present. Although it is not possible to detect a species with 100% certainty, using only observed site data will result in an under-detection bias. By surveying the same sites multiple times, the detection survey history can be used to estimate a detection probability,  $\rho$ , from the number of sites that are occupied,  $\Psi$ , and adjust the estimate for those survey occasions when the species is possibly present but was not detected.

The model  $\Psi(\cdot)\hat{\rho}(\cdot)$  was used for computations comparing the detection probability and occupancy estimates for the 2 trap types and 2 fruit fly species. This model assumes that the detection probability remained the same, and that the probability of occupancy was the same

for all sites for each wk over the 3 wk period. In the case of the sterile *C. capitata*, the weekly aerial release rate was constant for all sites and therefore it was assumed that constant numbers of flies were present at all sites. Although, this was obviously not the case for the naturally occurring *A. suspensa* population, we felt it safe to assume that no significant population changes occurred during the 3 wk period that the survey was conducted that would change site occupancy or detection. Therefore, this model was used to compare parameter estimates for the 2 trap designs and evaluate their effectiveness for each species under the same set of assumptions.

#### Statistical Analysis

For the test conducted in guava, effect of treatment was analyzed by ANOVA (Proc GLM, SAS Institute 2000) followed by Tukey's mean separation ( $P = 0.05$ ) for significant ANOVAs. Data were  $\log(x + 1)$  transformed prior to analysis to satisfy conditions of equal variance (Box et al. 1978), non-transformed means  $\pm$  standard deviations are presented. For the test conducted under sterile male *C. capitata* release, data were analyzed using an unequal variance  $t$  test (JMP 10, SAS Institute 2012). For a comparison at 2 levels, the unequal variance  $t$ -test is equivalent to the Welch ANOVA (Welch 1951). Data were  $\log(x + 1)$  transformed prior to analysis to satisfy conditions of equal variance (Box et al. 1978), and a standard  $F$  test for unequal variances was performed. The non-transformed means  $\pm$  standard deviations are presented. Separate analyses were conducted for each species and sex.

Following, Bailey et al. (2004), trap site estimates of  $\hat{\Psi}(\cdot)$  and  $\hat{\rho}(\cdot)$  were each used as the response variable for conducting separate ANOVAs to compare the main effects and interaction of trap type, trap site, and species using the model

error term for a least squares fit (JMP 10, SAS Institute 2012). A Student's  $t$  test was used to compare each paired effect.

#### RESULTS AND DISCUSSION

Type of trap and lure affected capture of *A. suspensa* in the test conducted in guava (Table 1). For both lures, there was higher capture in Multilure traps than in Phase 4 traps, and this was true for both males and females. There was higher capture of both sexes with two-component than three-component BioLure in Multilure traps, and this was also true for female capture in Phase 4 traps. The same pattern was observed for all sample periods within the field test. As has been observed in previous research, the highest capture throughout the study was in Multilure traps baited with the two-component BioLure.

The number of *A. suspensa* captured in the test conducted in the urban area was much lower than in the test conducted in guava, and there was again much higher capture of both females and males in the Multilure traps (Table 2). Similar results were obtained with number of sterile *C. capitata* captured in the 2 trap types. The lowest ratio of flies captured in Multilure traps versus Phase 4 traps was 5:1 and was obtained with sterile *C. capitata* capture. The same ratio of *A. suspensa* capture in the 2 traps baited with the same lure ranged from ~10:1 to ~100:1, indicating poorer relative capture of this species.

The percent of traps that are positive for fruit fly capture is often used as an indicator of how well a trap performs for detection in area-wide fruit fly detection programs. In this case, the observed trap site occupancy  $\Psi(\text{obs})$  for both trap types is used along with maximum likelihood estimates for occupancy  $\hat{\Psi}(\cdot)$  and detection probability  $\hat{\rho}(\cdot)$  for evaluation of each trap type and with regard to the 2 species (Table 3). In general, all of the estimates indicated an acceptable level

TABLE 1. MEAN ( $\pm$  STANDARD DEVIATION) NUMBER OF *ANASTREPHA SUSPENS*A CAPTURED PER TRAP PER DAY IN TRAPS BAITED WITH TWO- OR THREE-COMPONENT BIO LURE (AMMONIUM ACETATE [AA], PUTRESCINE [PU] WITH OR WITHOUT TRIMETHYLAMINE [TMA]) IN FIELD TESTS CONDUCTED IN A GUAVA ORCHARD, HOMESTEAD, FLORIDA ( $N = 5$ ).

Trap <sup>1</sup>	Lure	Females <sup>2</sup>	Males
Multilure	AA + Pu	53.2 $\pm$ 12.5 a	19.5 $\pm$ 3.6 a
Multilure	AA + Pu + TMA	25.2 $\pm$ 5.9 b	9.4 $\pm$ 2.2 b
Phase 4 trap	AA + Pu	1.5 $\pm$ 0.5 c	0.4 $\pm$ 0.2 c
Phase 4 trap	AA + Pu + TMA	0.4 $\pm$ 0.2 d	0.1 $\pm$ 0.1 c
$F_{3,12}$		740.07	809.37
$P$		<0.0001	<0.0001

<sup>1</sup>Multilure traps contained 300 mL 10% propylene glycol solution and Phase 4 traps had yellow sticky inserts to retain attracted flies.

<sup>2</sup>Means followed by the same letter within a column are not significantly different (Tukey's mean separation test on  $\log[x + 1]$ -transformed data, non-transformed mean  $\pm$  standard deviation presented).

TABLE 2. MEAN ( $\pm$  STANDARD DEVIATION) NUMBER OF *ANASTREPHA SUSPENS*A AND STERILE RELEASED *CERATITIS CAPITATA* CAPTURED PER TRAP PER DAY IN TRAPS BAITED WITH THREE-COMPONENT BIOLOURE (AMMONIUM ACETATE, PUTRESCINE AND TRIMETHYLAMINE) IN TESTS CONDUCTED IN URBAN AREAS OF BOCA RATON, FL ( $N = 100$ ).

Trap <sup>1</sup>	<i>A. suspensa</i> females Flies per trap per day	<i>A. suspensa</i> males Flies per trap per day	<i>C. capitata</i> sterile males Flies per trap per day
Multilure trap	3.0 $\pm$ 7.2	0.4 $\pm$ 1.14	0.5 $\pm$ 0.63
Phase 4 trap	0.08 $\pm$ 0.19	0.03 $\pm$ 0.10	0.1 $\pm$ 0.21
<i>t</i>	-6.022	-4.414	-5.262
<i>df</i>	103.4	105.9	140.7
<i>P</i>	< 0.0001	< 0.0001	< 0.0001

<sup>1</sup>Multilure traps contained 300 mL 10% propylene glycol solution and Phase 4 traps had yellow sticky inserts to retain attracted flies.

of precision in that the SE for each estimate/estimate was less than 25%. The observed trap site occupancy  $\Psi(\text{obs})$  for both trap types combined  $\Psi(\text{min})$  is used as the minimum number of sites known to be occupied with empirical certainty. The observed trap site occupancy for each trap type  $\Psi(\text{obs})$  was lower than the combined trap occupancy  $\Psi(\text{min})$  and the observed site occupancy probability  $\Psi(\text{obs})$  was consistently lower than the estimated occupancy  $\hat{\Psi}(\cdot)$  for both traps for both species, as well. These data illustrate the under-estimate bias that exists, which ecologists call the “naïve occupancy estimate,” when using only the observed presence data (MacKenzie et al. 2002). In the case of Multilure traps, the occupancy estimate  $\hat{\Psi}(\cdot)$  was very similar to the combined trap occupancy  $\Psi(\text{min})$ . For the Phase 4 trap, only the occupancy estimate  $\hat{\Psi}(\cdot)$  for *C. capitata* approached that of the combined trap occupancy  $\Psi(\text{min})$ . In both trap types, the occupancy estimates  $\hat{\Psi}(\cdot)$  were considerably higher for sterile *C. capitata* than for wild *A. suspensa*. This would be expected since the sterile flies were released by aircraft at a standard weekly rate over all 100 sites surveyed. Therefore, we would assume that there would be at least one sterile *C. capitata* detected in each survey area. Indeed the occupancy estimates approached 100% for both the Multilure and the Phase 4 trap at 93% and 97%, respectively. In the case of *A. suspensa*, both the occupancy and the detection probabilities were consistently lower than the Multilure trap. This could support the observation made earlier that there is a higher relative effectiveness of the Phase 4 trap for sterile *C. capitata* than for *A. suspensa*. However, the occupancy estimates for *A. suspensa* with the Phase 4 traps are very suspect in that they are less than half of that seen in the Multilure traps as well as the observed minimum combined trap site occupancy  $\Psi(\text{min}) \pm \text{SE}$  (see notes: Table 3). Therefore, both the occupancy estimates  $\hat{\Psi}(\cdot)$  and the detection probability estimates  $\hat{\rho}(\cdot)$  are not considered valid with respect to *A. suspensa* for the Phase 4 trap. A combination of 2 factors may have contributed to the lower occu-

pancy observed for *A. suspensa*. One apparent difference between the 2 species with regard to site occupancy and detection as surveyed in this test is that of aggregation or clumped distributions. The distributions of natural fruit fly populations have been shown to be aggregated (Meats 2007; Puche et al. 2005; Zalucki et al. 1984). The sterile *C. capitata* males surveyed in this test were artificially distributed evenly on a weekly basis and as already noted, had very high site occupancy estimates  $\hat{\Psi}(\cdot)$ . Whereas, the site occupancy estimates for the naturally occurring *A. suspensa* population, as detected in the three-component Multilure traps, were much lower. This reveals a potential second flaw for making a comparison of site occupancy estimates of the 2 trap designs using the three-component lure rather than the two-component lure for detection of *A. suspensa*. As was confirmed herein in the test in guava, *A. suspensa* is not as attracted with the three-component lure as it is with the two-component lure, putrescine and ammonium acetate. Therefore, in the case of *A. suspensa*, the Phase 4 trap might have had different site occupancy and detection estimates, if the two-component lure was used rather than the three-component lure.

Tables 4 and 5 give the results of ANOVA effects tests on the occupancy and detection probability estimates, respectively. Occupancy estimates varied significantly with trap type, trap site, and species. There was a significant trap x species interaction for occupancy estimate as well, reflecting the comparatively higher occupancy estimates for *C. capitata* (relative to *A. suspensa*) for the Phase 4 traps than the Multilure traps. Even though there were significant differences in specific site occupancy estimates, the trap site interaction was not different as would be expected for paired trap types at each site. From this, site occupancy estimates would also be expected to be fairly stable for each specific species regardless of the trap type used. In the case of *A. suspensa*, the Multilure trap would appear to have a greater potential for detection than Phase 4 trap. The species had a significant effect on detection prob-

TABLE 3. SITE OCCUPANCY AND DETECTION PARAMETER ESTIMATES FOR MULTILURE AND PHASE 4 TRAP SITES ON 3 SEPARATE SURVEY OCCASIONS ( $N = 100$ ).

Species	Sex	Multilure (wet)			Phase IV (dry)			
		$\Psi(\text{min})$	$\Psi(\text{obs})$	$\hat{\Psi}(\cdot) [1 \text{ SE}]$	$\hat{\rho}(\cdot) [1 \text{ SE}]$	$\Psi(\text{obs})$	$\hat{\Psi}(\cdot) [1 \text{ SE}]$	$\hat{\rho}(\cdot) [1 \text{ SE}]$
<i>C. capitata</i>	Male	0.88	0.82	0.93 [0.05]	0.50 [0.04]	0.66	0.97 [0.12]	0.32 [0.05]
<i>A. suspensa</i>	Male	0.45	0.43	0.46 [0.05]	0.59 [0.05]	<b>0.21</b>	<b>0.30 [0.07]</b>	0.34 [0.08]
<i>A. suspensa</i>	Female	0.64	0.62	0.65 [0.05]	0.66 [0.04]	<b>0.28</b>	<b>0.32 [0.05]</b>	0.52 [0.07]
<i>A. suspensa</i>	both	0.68	0.66	0.71 [0.05]	0.59 [0.04]	<b>0.32</b>	<b>0.35 [0.05]</b>	0.55 [0.06]

Notes: Results are for the *ts*/Mediterranean fruit fly sterile release, *Ceratitis capitata*, and the native *Anastrepha suspensa* population in Florida. The observed site occupancy is given for each trap type ( $\Psi(\text{obs})$ ). The probability of occupancy estimates ( $\hat{\Psi}(\cdot)$ ) and detection probability estimates ( $\hat{\rho}(\cdot)$ ) are also given using maximum likelihood methods found with PRESENCE software (MacKenzie et al. 2002, 2003). The proportion of sites occupied with the Multilure and the Phase 4 traps combined is given as the minimum observed site occupancy ( $\Psi(\text{min})$ ). Boldface indicates poor occupancy estimates, where the estimated site occupancy interval ( $\hat{\Psi}(\cdot) \pm \text{SE}$ ) does not contain the minimum observed site occupancy ( $\Psi(\text{min})$ ).

abilities as well, but in this case the interaction term was not significant. However, as pointed out in Table 3, the site occupancy estimates for *A. suspensa* in the Phase 4 trap are not considered to be valid estimates and should not be used to make a comparison.

A trap-lure comparison study conducted for *C. capitata* in coffee in Hawaii found that the Multilure trap baited with the three-component BioLure significantly outperformed Multilure traps baited with two-component BioLure or Torula yeast, and Phase 4 traps baited with both two- and three-component BioLure. However, the Phase 4 trap capture was not significantly different from the other trap-lure combinations in the study (T. Shelly, unpublished data). Studies conducted in low elevation coffee (600 - 650 m) in Guatemala found equal capture of sterile males in Phase 4 traps baited with three-component BioLure and trimedlure-baited Jackson traps (Jeronimo et al. 1999). In other tests conducted in Guatemala but in high elevation coffee (1000 - 1,800 m), trimedlure-baited Jackson traps captured more male *C. capitata* than Phase 4 traps baited with three component BioLure with a ratio of 6.5:1 sterile males and 1.7:1 wild males captured (Midgarden et al. 2004). Ratios of sterile males captured in trimedlure-baited Jackson traps versus Multilure traps baited with three-component BioLure are typically 7:1 in releases under the Cooperative Fruit Fly Detection Program in Florida (D.E.D., unpublished data).

Research conducted in Mexico compared captures of wild *C. capitata*, *A. ludens*, and *A. obliqua* in coffee and in fruit orchards (Montoya et al. 1999). These tests evaluated the two- and three-component BioLures in open-bottom dry traps and plastic McPhail traps, and trimedlure-baited Jackson traps among other trap-lure combinations tested. Populations were extremely low in these tests, with less than 1.0 fly per trap per day for female and male *C. capitata* and less than 0.1 fly per trap per day for either *Anastrepha* species. Except for a few trials in which the highest captures of male *C. capitata* were in Jackson traps baited with trimedlure, there were no differences in any species or sex among all trap and lure combinations. However, in a similar test in Mexico with *A. ludens* on oranges and pears, and *A. obliqua* on mango, Thomas (2010) found that there was a stark difference between the two-component BioLure in a Multilure trap compared to a Phase 4 dry trap. The Phase 4 trap failed to capture any flies for eight wk of the twelve wk test and had a combined 1% of total *A. ludens* captured, whereas the Multilure trap captured the most flies, with 75% more than the closest competitor. Again, with *A. obliqua*, the Multilure trap with two-component BioLure had the best results, and the Phase 4 had the worst of any trap-lure combination tested.

TABLE 4. EFFECTS TEST OF TRAP TYPE, TRAP SITE, AND SPECIES ON THE PROBABILITY OF OCCUPANCY ESTIMATE  $\hat{\Psi}(\cdot)$ , USING ANOVA FOR MODEL,  $\hat{\Psi}(\cdot)\hat{\rho}(\cdot)$  ON THE COMBINED DATA ( $N = 600$ ).

Source	df	SS	F	P
Trap	1	0.4789	5.0092	0.0258
Species	1	35.9332	375.8347	< 0.0001
Site	99	29.3381	3.0995	< 0.0001
Trap $\times$ Species	1	2.8496	29.8047	< 0.0001
Trap $\times$ Site	99	8.6019	0.9088	0.7135
Error	398	38.0524		

The summary of the findings reviewed here does not explain the inconsistent results regarding the capture of *C. capitata* reported with the Phase 4 trap at various locations, elevations, and hosts. The Phase 4 trap is either performing very poorly or, on average, equal with trimedlure, Bio-Lure, and torula yeast in detection of *C. capitata*, while it has never been reported to be effective with various species of *Anastrepha*. The effect of trap design on the ability to capture and retain fruit flies once they are attracted may be related to some environmental or host behavioral differences in fruit fly response. Prokopy et al. (1996) found in a field cage behavioral observations that there was a significant difference in the percent of protein fed and starved flies that entered traps. However, there was no difference in the percent of mature female or male *C. capitata* that entered a glass McPhail trap baited with 9% Nulure/5% borax and an early version of the Phase 4 trap baited with the two-component lure, ammonium acetate and putrescine. This causes us to speculate that the large difference that we observed between the capture of *A. suspensa* in the Multilure and Phase 4 traps may have more to do with a combination of the effectiveness of the lure and the ability of the Phase 4 trap to retain the attracted flies. The total non-sticky surface area that is available for flies to alight when attracted to the open bottom Phase 4 trap is 5.2 times greater than the 7.6 by 12.7 cm surface area of the yellow sticky panel coated on both sides, which is suspended inside the trap. Without the aid of a toxicant, the attracted flies are at much more liberty to escape without getting retained on the

yellow sticky panel than flies that enter a Multilure trap with a single inverted opening design. If that is the case, it is possible that the Phase 4 trap would have performed better for *A. suspensa* in the urban test if the two-component lure rather than the three-component lure had been used. That in turn might have resulted in slightly better site occupancy and detection probability estimates for *A. suspensa* that could have been valid for making trap comparisons.

Large scale area-wide deployment of fruit fly detection traps is costly in the trap design and materials, the attractants used, and in the time and effort required in routinely servicing them. Although a simpler and cheaper trap such as the dry Phase 4 would be a welcome relief to any large scale area-wide detection programs, it must perform effectively. The Phase 4 trap did perform comparably with the Multilure trap in terms of site occupancy estimates and detection probabilities, even though it caught 5 times fewer total *C. capitata*. If this observation was found to be consistent, then the Phase 4 trap might have a place in program cost savings both in terms of the relative low cost of the trap and a reduction in the number of flies that have to be screened without sacrificing detection probability. However, with regard to *A. suspensa*, the observations in these trials and many others cited here for other species of *Anastrepha*, it cannot currently be recommended for use under trapping conditions in the Cooperative Fruit Fly Detection Program in Florida. We believe that with further development lures that can improve both attraction and retention of flies at the trap and possibly an increase in the

TABLE 5. EFFECTS TEST OF TRAP TYPE, TRAP SITE, AND SPECIES ON THE PROBABILITY OF DETECTION ESTIMATE  $\hat{\rho}(\cdot)$ , USING ANOVA FOR MODEL,  $\hat{\Psi}(\cdot)\hat{\rho}(\cdot)$  ON THE COMBINED DATA ( $N = 600$ ).

Source	df	SS	F	P
Trap	1	0.0526	11.2397	0.0009
Species	1	1.8408	392.8427	< 0.0001
Site	99	$9.28 \times 10^{-30}$	0.0000	1
Trap $\times$ Species	1	0.0075	1.6005	< 0.2066
Trap $\times$ Site	99	$8.24 \times 10^{-28}$	0.0000	1
Error	398	1.8650		

capture surface, the Phase 4 trap might have the potential to reduce large scale area-wide fruit fly detection costs without greatly sacrificing the probability of detection. To our knowledge this is the first time that the software PRESENCE has been used to evaluate fruit fly detection probabilities. It may find further application in future area-wide fruit fly detection and trap evaluation programs.

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