

Life table assay of field-caught Mediterranean fruit flies, *Ceratitis capitata*, reveals age bias

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Abstract

Though traps are used widely to sample phytophagous insects for research or management purposes, and recently in aging research, possible bias stemming from differential response of individuals of various ages to traps has never been examined. In this paper, we tested the response of *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) males and females of four ages (spanning from 1 to 40 days) to McPhail-type traps baited with a synthetic food attractant in field cages and found that the probability of trapping was significantly influenced by age. The type of food on which flies were maintained before testing (sugar or protein) also had a strong effect and interacted with age. In another experiment, we collected wild *C. capitata* adults of unknown age using 1–3 methods and then reared them in the laboratory until death. The survival schedules of these flies were subsequently used in a life table assay to infer their age at the time of capture. Results showed that on a single sampling date, males captured in traps baited with a food attractant were younger compared with males aspirated from fruiting host trees, or males captured in traps baited with a sex attractant. Likewise, females captured in food-baited traps were younger compared with aspirated females. In addition to providing the first evidence of age-dependent sampling bias for a phytophagous insect species, this paper also provides a novel approach to estimate the differences in the age composition of samples collected with different techniques. These findings are of utmost importance for several categories of insects, medically important groups notwithstanding.

Introduction

One of the most important problems in ecology and biodemography is understanding aging in the wild. Insect traps are commonly used to draw a sample from wild populations and then use information gained from this sample to infer age-structure and other population parameters that apply to the entire population (Southwood, 1978; Pedigo & Buntin, 1994; Müller et al., 2007;

Carey et al., 2008). Traps that lure and capture actively-moving insects rely on visual, chemical, or acoustic cues that are normally used by insects to locate resources for feeding, members of the opposite sex for mating, and oviposition sites for egg laying (Epsky et al., 2004). These cues are perceived by one or more of the senses and affect insect orientation and other behaviors such that capture by the trap is facilitated. However, an insect's age can affect foraging behavior and response to cues (e.g., Seabrook et al., 1979; Jang, 1995; Kendra et al., 2005). As a result, random samples that represent all the age classes in a population may be very difficult to obtain with traps or other collection techniques that manipulate the behavior of insects.

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Despite the enormous amount of resources that has been invested in evaluating various insect trapping techniques and despite the importance of interpreting trap capture, it is remarkable that the entomological literature contains only a handful of studies that acknowledge or investigate potential age-dependent trap bias (e.g., Owaga & Challier, 1985; Van Sickle & Phelps, 1988; Wall et al., 1991), none of which concerns phytophagous insects. Researchers often assume that the probability of capturing insects is independent of age. Although in certain cases this assumption is justified, there are situations where an understanding of age bias in sampling is important. For example, if the probability of capturing newly emerged individuals is low, then the likelihood of detecting populations consisting of mostly young individuals may be small. Similarly, programs for monitoring insect disease vectors (e.g., mosquitoes) may yield erroneous results and thus alter policy if certain age classes are more likely to be detected than others.

One reason for this lack of studies is that there are methodological constraints for accurately estimating the age of insects derived from the field. The limited number of age determination methods provide unreliable results, particularly when used to estimate the ages of older members in a population. These methods are based on follicular relics in females (Tyndale-Biscoe, 1984), the accumulation of pteridines in insect eye capsules (Mail et al., 1983; Lehane et al., 1986), transcriptional profiling (Cook et al., 2006), or the progressive layering of hydrocarbons in insect cuticles (Desena et al., 1999; Gerade et al., 2004). Moreover, these methods can essentially be applied only to a few insect species of medical and veterinary importance. In fruit-infesting fruit flies (Tephritidae), as well as several other categories of insects that include important agricultural pests, the only age class that can be recognized with certainty is that of very young adults, which do not yet have mature oocytes or sperm (Fletcher et al., 1978; Kendra et al., 2006).

A newly introduced concept which relies on trapping but circumvents the need for age determination techniques is that of residual demography (Müller et al., 2004, 2007; Vaupel, 2009). Individuals are trapped from the wild population at unknown ages and the resulting captive cohort is reared out in the laboratory until death. Using the residual life span of these flies one can infer the age-structure of the wild population. This method renders trapping a powerful tool for understanding aging in the wild, but requires unbiased trapping that provides random samples. A recent study by Carey et al. (2008) aimed at studying aging in the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), a worldwide pest of major economic importance, the principles of residual demography were applied on the captive life spans of over 4 000 wild-trapped individuals. This study provided several important

findings, such as that major changes in population age-structure occur in wild *C. capitata* populations of both sexes and that middle-aged (3–4 weeks) and older individuals are common throughout much of the field season in central Greece. Spring *C. capitata* populations tended to be older and late summer and fall populations tended to be younger. In Carey et al.'s (2008) study, the possibility that the trapping method used was biased towards capturing certain age classes over other age classes was acknowledged as a potential source of bias.

This follow-up study was aimed at estimating the extent to which capture methods influence the age composition of sampled populations of *C. capitata*. We employed the same trapping method used by Müller et al. (2007) and Carey et al. (2008), which consists of McPhail-type traps (McPhail, 1939) baited with a food-based synthetic attractant. This trap has various versions and is a standard tool for capturing numbers of *C. capitata* and other fruit flies (IAEA, 2003). The trap can be used as a direct control tool to prevent or suppress population build up of *C. capitata* by mass trapping, but is also used widely as an aid for other management purposes. Baited with the appropriate lures, it captures predominately females of *C. capitata* (Heath et al., 1997; Epsky et al., 1999; Katsoyannos et al., 1999) and is particularly sensitive in detecting low populations (Papadopoulos et al., 2001). Mostly these traits have rendered it a valuable tool in several places in the world where the sterile insect technique (Knipling, 1955) and other important methods for controlling or eradicating this insect are applied. These methods are most effective when applied against small target populations (Dowell et al., 1999). Populations resulting from surviving overwintering larvae escape detection by other trapping systems and may remain unnoticed for long periods following emergence (Carey, 1996; Papadopoulos et al., 2001).

We initially examined potential age-dependent bias under semi-natural conditions by counting captures of *C. capitata* males and females of various known ages in McPhail-type traps suspended from the ceiling of a field cage. To gain further evidence of age-dependent bias, in a second experiment we collected wild *C. capitata* adults of unknown age using various methods and then reared them out in the laboratory until death. The survival schedules of these flies were subsequently used in a life table assay to determine whether different collection methods selected for different age categories of flies.

Materials and methods

Field cage experiment

The flies used in this experiment were retrieved from naturally infested figs (*Ficus carica* L.; Moraceae) and were kept

in wire-screened cages (30 × 30 × 30 cm) in the laboratory (25 ± 2 °C) from emergence until they reached test age. Each cage contained 100–200 individuals of both sexes, water, and one of two different types of food. The experiment involved four age groups of *C. capitata* males and females, corresponding to different stages of the reproductive cycle and life history (Carey, 1984; Papadopoulos et al., 2002, 2004). These age groups were flies of 1–5 days (young immature), 10–15 days (young mature), 20–25 days (middle-age mature), and 35–40 days (older mature). Flies were fed either only sugar or a protein diet, consisting of a mixture of yeast hydrolysate (ICN Biomedicals, Irvine, CA, USA) and sugar in a ratio of 1:4. The tests were conducted in October 2005 in six cylindrical (2 m high, 2.6 m diameter), plastic-screen field cages, set out in an open area at the University of Volos, Greece. Each housed a potted non-host tree (*Ficus benjamina* L.), which was washed thoroughly to remove possible food sources for the flies. To conduct a test, 50 males and 50 females of a given age- and food-group were introduced into a field cage. Half an hour later, a dry Multilure[®] trap (Better World Manufacturing, Fresno, CA, USA) was suspended by a 10-cm-long wire from the ceiling of each cage, at a height of about 1.8 m above ground. The trap was baited with the two components ammonium acetate and trimethylamine of the commercially available food-based synthetic attractant Biolure[®] (Suterra, Bend, OR, USA), which are highly effective for capturing *C. capitata* when used in combination (Katsoyannos et al., 1999). After 1 h, the number of flies that had entered the trap was counted and this formed a replicate. Before a new replicate was run, all flies in the field cage were replaced with new ones. Tests were conducted between 09:00 and 17:00 hours over the course of 1 week. A light to moderate wind blew through the cages during most days of the study and temperature ranged between 23 and 25 °C. For each age group we ran 5–7 replicates.

Let μ denote the corresponding trapping probability, $\mu = P(y = 1)$ and assume a binomial variance function $\text{var}(y) = \Phi\mu(1 - \mu)$, where Φ is the overdispersion parameter. This model is fitted by a quasi-likelihood approach, aiming to maximize the quasi-likelihood, which is defined as follows:

$$Q(\mu, y) = \sum_{i=1}^n \left[y_i \log \frac{\mu_i}{1 - \mu_i} + \log(1 - \mu_i) \right]$$

where n is the total number of flies tested, 5 100, and $\mu_i = \frac{\exp(\beta'x_i)}{1 + \exp(\beta'x_i)}$, or equivalently, $\log \frac{\mu_i}{1 - \mu_i} = \beta'x_i$, $i = 1, \dots, n$. Here x_i is the predictor or covariate vector for the i th observation y_i . The predictors include type of food (sugar = 1, protein = 0, i.e., protein is baseline); age group, with baseline group 1–5, corresponding to three

indicator variables for age groups 10–15, 20–25, and 35–45, respectively; and sex (male = 1, female = 0, i.e., female is baseline).

Open field experiment

The main aim of the second part of the study was to determine whether different collection methods selected for different age categories of flies. One of these methods (the Multilure trap baited with two-component synthetic food attractant, as described above) was also used in two recent studies investigating aging in the wild (Müller et al., 2007; Carey et al., 2008). Using 15 such traps, free-ranging adults were collected during the 2006 field season in a citrus orchard on Chios Island (38°21'N, 26°08'E), Greece. The orchard is located within the Campos area, which is almost exclusively occupied by citrus cultivations and has few other hosts (Katsoyannos et al., 1998). These 1-day collections were made on six sampling dates (21 June, 9 and 23 July, 6 August, and 2 and 17 October) in order to follow changes in the age-structure of the population. To gain evidence of age-dependent trapping bias by comparing different sampling techniques, on the last four of these dates, flies (males only) were also collected with three Multilure traps baited with the male-specific sexual attractant Trimedlure (AgriSense, Fresno, CA, USA). For the same purpose, on late afternoon of 21 July, a single collection was made of flies observed feeding on natural wounds on fruits of two plum trees, *Prunus domestica* L. (Rosaceae), by aspiration by two persons. We assumed that this collection would yield a representative sample of the population, because adults require constant intake of food for maintenance and they are mainly present at the feeding arenas at this time of day (Hendrichs et al., 1991). The 1–3 cohorts captured on the various dates were air-shipped alive for same-day arrival to the University of Thessaloniki, Greece. There they were placed individually in 6 × 9.5 × 12 cm transparent plastic cages (inverted plastic cups of 0.4 l capacity), provided with water and adult food (4:1 ratio of sugar and yeast hydrolysate) in a laboratory room at 25 ± 2 °C. Survival of both sexes was monitored daily. Stress-related mortality (e.g., due to transport and transfer) was extremely low (<<1% during the first 48 h post-capture) (Carey et al., 2008).

The post-capture survival schedules of these flies were used to infer changes in age-structure during the season as well as tendencies of the collection methods to select for different age categories of flies. The basic assumption was that relative differences in the survival rates of flies captured using different techniques would reflect relative differences in age composition and/or frailty of the captured flies. Data were analyzed with a two-way analysis of variance. Factors were 'capture date' and 'trap type'. Data were

transferred age to $\sqrt{\text{age}}$ for all inferential analysis. In the analysis, 'aspiration' was used as the baseline method and '17 October' as the baseline date.

Results

Field cage demonstration of age-dependent trapping bias

A total of 1 081 flies or slightly over 21% of all individuals were captured using traps. The percentages of responding males and females were comparable (48.2 vs. 51.8%, respectively). Unlike sex, type of food had a strong effect on captures (Figure 1). Overall, flies fed only on sugar responded four-fold stronger than flies fed on protein (79.5 vs. 20.5%, respectively).

Fitting all predictors and their interactions in a preliminary model, we reduced some insignificant predictors and retained those interactions between food type and age groups that are significant (their respective main effects

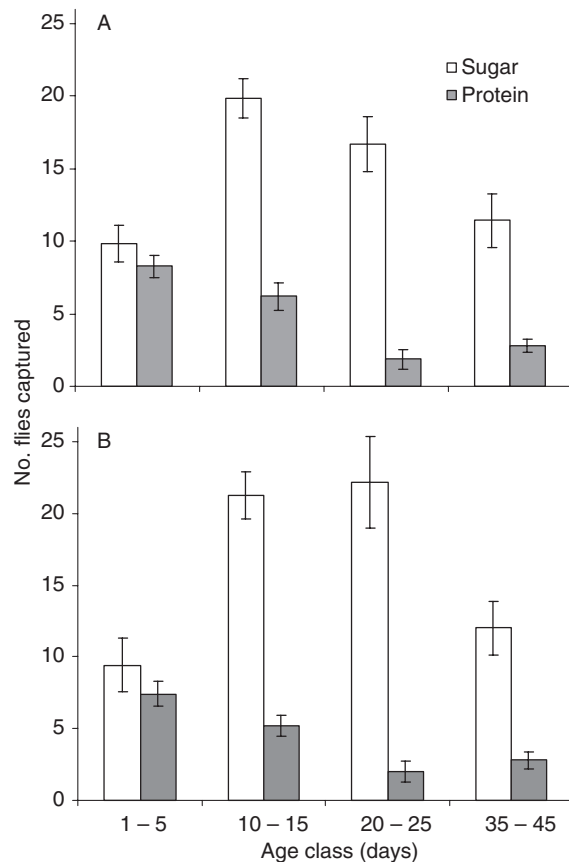


Figure 1 Differential response (mean \pm SE) of wild *Ceratitis capitata* (A) males or (B) females of four ages and two feeding states to a McPhail-type trap baited with a two-component synthetic food attractant in a field cage. Each bioassay involved 50 flies from either sex and the duration was 1 h. The number of replicates corresponding to each bar (treatment) was 5–7.

are also included). Main effects and the interactions between food and age kept in the model effect are given in Table 1. Here factor 'age 10–15', corresponds to the indicator of age group 10–15, it is equal to 1 if the fly belongs to age group 10–15 and 0 otherwise – analogously for ages 20–25 and 35–40. Likewise, factor 'sugar*age 10–15' corresponds to the interaction term between sugar group and age group 10–15, and is equal to 1 if the fly belongs to the sugar group and the age group 10–15 – analogously for the other interactions. In this final model, age 20–25, age 35–45, and the interactions between type of food and each age group are significant. The estimated overdispersion parameter is 1.77. This quasi-logit model can be summarized as follows, for the linear predictor $\eta_i = \log \left[\frac{\mu_i}{1-\mu_i} \right]$, where μ_i is the trapping probability for the i -th fly:

$$\begin{aligned} \eta_i = & -1.630 + 0.248I_i(\text{sugar}) \\ & - 0.371I_i(\text{age}10 \sim 15) \\ & - 1.376I_i(\text{age}20 \sim 25) \\ & - 1.145I_i(\text{age}35 \sim 45) \\ & - 0.102I_i(\text{male}) \\ & + 1.445I_i(\text{age}10 \sim 15)I_i(\text{sugar}) \\ & + 2.354I_i(\text{age}20 \sim 25)I_i(\text{sugar}) \\ & + 1.393I_i(\text{age}35 \sim 45)I_i(\text{sugar}), \end{aligned}$$

where $I_i(\cdot) = 1$ when the i th fly belongs to the group specified in parentheses, and 0 otherwise.

Table 1 Main effects and interaction between food type and age for the probability of capture of *Ceratitis capitata* males and females of various ages in Multilure traps baited with a two-component synthetic food attractant, in field cages

Coefficients/factors	Estimate \pm SE	t value	Pr(> t)
Intercept	-1.630 \pm 0.146	-11.162	<0.001
Sex (males)	-0.102 \pm 0.096	-1.058	0.293
Food (sugar)	0.248 \pm 0.188	1.318	0.191
Age 10–15	-0.371 \pm 0.233	-1.592	0.115
Age 20–25	-1.376 \pm 0.297	-4.638	<0.001
Age 35–40	-1.145 \pm 0.294	-3.899	<0.001
Sugar*age 10–15	1.445 \pm 0.285	5.075	<0.001
Sugar*age 20–25	2.354 \pm 0.339	6.944	<0.001
Sugar*age 35–40	1.393 \pm 0.342	4.076	<0.001

The flies were obtained from wild hosts and, before testing, were maintained on two types of food in the laboratory. Each bioassay involved 50 flies from either sex and the number of replicates was 5–7 per treatment.

The model relates the covariates with the linear predictor η , such that the probability of capture $\mu = \exp(\eta) / [1 + \exp(\eta)]$. The larger η , the higher is the probability of capture; if $\eta = \beta_0$, the intercept, then one obtains the probability when all covariates are at their baseline levels (i.e., protein, female, or age group 5–10).

We can use odds ratios to demonstrate the effect of type of food in each age group and the age effect for each type of food. The odds ratio of the sugar group to the protein group is defined as

$$\frac{P(y = 1|\text{sugar})/[1 - P(y = 1|\text{sugar})]}{P(y = 1|\text{protein})/[1 - P(y = 1|\text{protein})]},$$

where $P(y = 1|\text{sugar})$ and $P(y = 1|\text{protein})$ are the trapping probabilities in the sugar and protein groups, respectively. The odds ratio of the sugar group to the protein group was 1.28, 5.44, 13.50, and 5.16 for the four age groups, respectively, which shows that the sugar method results in greater trapping probabilities in all age groups, especially in the 10–15, 20–25, and 35–45 age groups.

The odds ratio of an older age group, for example the 10–15 age group, to the youngest age group (baseline age group, 1–5) is defined as

$$\frac{P(y = 1|\text{age}10 \sim 15)/[1 - P(y = 1|\text{age}10 \sim 15)]}{P(y = 1|\text{age}1 \sim 5)/[1 - P(y = 1|\text{age}1 \sim 5)]},$$

where $P(y = 1|\text{age}10 \sim 15)$ and $P(y = 1|\text{age}1 \sim 5)$ are the trapping probabilities in age groups 10–15 and 1–5, respectively – similarly for other age groups. The calculated odds ratios were 2.93, 2.66, and 1.28 for the age groups 10–15, 20–25, and 35–45, respectively. This analysis shows that in the sugar group the middle age groups (10–15 and 20–25 days) were more responsive to the trap than the younger one (1–5 days) and the older one (35–45 days), whereas the older age group (35–45 days) was slightly more responsive than the younger age group (1–5 days). However, in the protein group, the younger age groups (1–5 and 10–15 days) were more responsive, where age group 1–5 days had the greatest trapping probability (odds ratios 0.69, 0.25, and 0.32 for age groups 10–15, 20–25, and 35–45, respectively).

Age differences in flies captured with different methods

During this study, a total of 1 677 *C. capitata* adults were collected in the citrus orchard. In males, the mean post-capture (remaining) survival times differed between capture dates by up to 28 days in Multilure traps baited with the two-component food attractant, and up to 37 days in traps baited with Trimedlure (Table 2). In females captured with the food attractant, the mean post-capture survival times differed by up to 28.4 days between capture dates. Differences between certain capture dates were statistically significant (Table 3), indicating changes in the age-structure of the population during the field season. Trimedlure captured twice as many flies as the food attractant, showing that it is a better lure for males in Multilure traps.

The results of trapping studies revealed that the average survival of males captured on 23 July with the food-baited

Multilure traps was 10.7 days longer (59.9 vs. 49.2 days) than with Trimedlure-baited traps, and 11.2 days longer (59.9 vs. 48.7 days) than with the baseline aspiration method (Table 2). The differences in post-capture survival times between food-baited traps and either of the other methods were significant (Table 3). Record life span was also longer, by 22 days (169 vs. 147 days) and 53 days (169 vs. 116 days), respectively. The survivorship curves of these three categories of males are given in Figure 2.

Based on our assumption that relative differences in the survival rates of captured flies reflect relative differences in age composition, these results suggest that on the day of collection the males that responded and were captured in Multilure-baited traps were younger than the males aspirated from the fruiting host trees or the males that were captured in Trimedlure-baited traps. On the other three collection dates, however, no significant differences were observed in the age of males captured in Multilure- or Trimedlure-baited traps (Table 3, Figure 3).

Regarding females, those captured on July 23 in the food-baited traps survived, on average, 8 days longer (49.1 vs. 41.1 days) than those that were aspirated from the host tree (Table 2). This difference in post-capture lifetime is significant and similar to that in males (Table 3, Figure 3). On the day of collection, therefore, females that were captured in Multilure-baited traps were younger than the aspirated females.

Discussion

The results of the present study show that one of the most widely used trapping systems for *C. capitata* (a variation of the food-based McPhail trap) is liable to a strong age-dependent bias. This bias stems from a differential capture probability of individuals of various ages and results in samples that may not reflect the actual age-structure of a wild population. The few previous studies that examined age-dependent trap biases concerned insects of medical and veterinary importance for which age estimation techniques are available (Owaga & Challier, 1985; Van Sickle & Phelps, 1988; Wall et al., 1991). In most other insect species, the ages of adults cannot be differentiated and, as a result, the study of age-dependent bias and other aspects of insect biology that depend on age have not advanced.

Our findings have important implications for understanding aging in the wild using the newly developed technique of residual demography (Müller et al., 2004, 2007; Carey et al., 2008; Vaupel, 2009). This technique does not depend on age determination methods, which are lacking for most insects, but uses instead the captive life span of individuals that are trapped at unknown ages and then reared out in the laboratory until death. This technique,

Table 2 Capture dates, numbers captured (n), and summary statistics [mean post-capture survival times \pm SD and maximum post-capture survival times (max)] for *Ceratitis capitata* flies captured with various methods during the 2006 field season on Chios Island, Greece

	Collection date	Multilure trap with a food attractant			Multilure trap with Trimedlure			Aspiration		
		n	Mean \pm SD (days)	Max (days)	n	Mean \pm SD (days)	Max (days)	n	Mean \pm SD (days)	Max (days)
Males	21 June	13	70.1 \pm 36.2	110	–	–	–	–	–	–
	9 July	50	63.8 \pm 33.6	172	–	–	–	–	–	–
	23 July	160	59.9 \pm 35.4	169	211	49.2 \pm 30.4	147	233	48.7 \pm 27.6	116
	26 August	21	42.1 \pm 32.4	115	80	43.8 \pm 32.1	113	–	–	–
	2 October	18	68.3 \pm 41.2	154	44	80.8 \pm 43.5	154	–	–	–
	17 October	28	54.5 \pm 22.2	93	142	58.9 \pm 28.6	124	–	–	–
Females	21 June	75	46.9 \pm 23.0	118	–	–	–	–	–	–
	9 July	50	59.3 \pm 29.1	149	–	–	–	–	–	–
	23 July	268	49.1 \pm 30.7	127	–	–	–	66	41.1 \pm 28.4	125
	26 August	69	43.7 \pm 22.2	105	–	–	–	–	–	–
	2 October	125	72.1 \pm 40.4	173	–	–	–	–	–	–
	17 October	24	56.5 \pm 21.3	105	–	–	–	–	–	–

Table 3 Effects of date and trap type on post-capture life spans of male and female *Ceratitis capitata* captured with various methods during the 2006 field season on Chios Island, Greece

Coefficients/factors	Estimate \pm SE	t-value	Pr(> t)
Males			
Intercept	7.143 \pm 0.266	26.833	<0.001
Method food attractant	0.481 \pm 0.221	2.172	<0.05
Method Trimedlure	0.182 \pm 0.210	0.870	0.384
21 June	0.209 \pm 0.682	0.306	0.760
9 July	0.070 \pm 0.404	0.174	0.862
23 July	–0.504 \pm 0.219	–2.296	<0.05
26 August	–1.367 \pm 0.289	–4.723	<0.001
2 October	0.964 \pm 0.342	2.814	<0.005
Females			
Intercept	6.776 \pm 0.529	12.807	<0.001
Method food attractant	0.614 \pm 0.296	2.077	<0.05
21 June	–0.805 \pm 0.504	–1.596	0.111
9 July	0.101 \pm 0.534	0.189	0.850
23 July	–0.742 \pm 0.458	–1.620	0.106
26 August	–1.020 \pm 0.510	–2.002	<0.05
2 October	0.720 \pm 0.479	1.501	0.134

Survival times have been \sqrt{x} -transformed to obtain normality; the two factors are date of capture and trap type, with baselines aspiration method and 17 October; the numbers of flies captured on each collection date are given in Table 2; the intercept denotes the baseline post-capture life span to which the effects on post-capture life span by the various conditions are added; the effects are overall significant (F-test: $P < 0.001$).

however, is seriously compromised if the trapping methods used are biased towards capturing certain age classes over others. To infer age-related population parameters that apply to the entire population we need random samples or, alternatively, knowledge of the variation in probability of being sampled among age groups. In the latter case,

compensatory steps can be taken during the analysis and interpretation of the data so that a more accurate estimate of population age-structure may be achieved (Spradbery & Vogt, 1993; Binns et al., 2000). The approaches for estimating age-dependent bias presented here can help improve the accuracy of the residual demography tool.

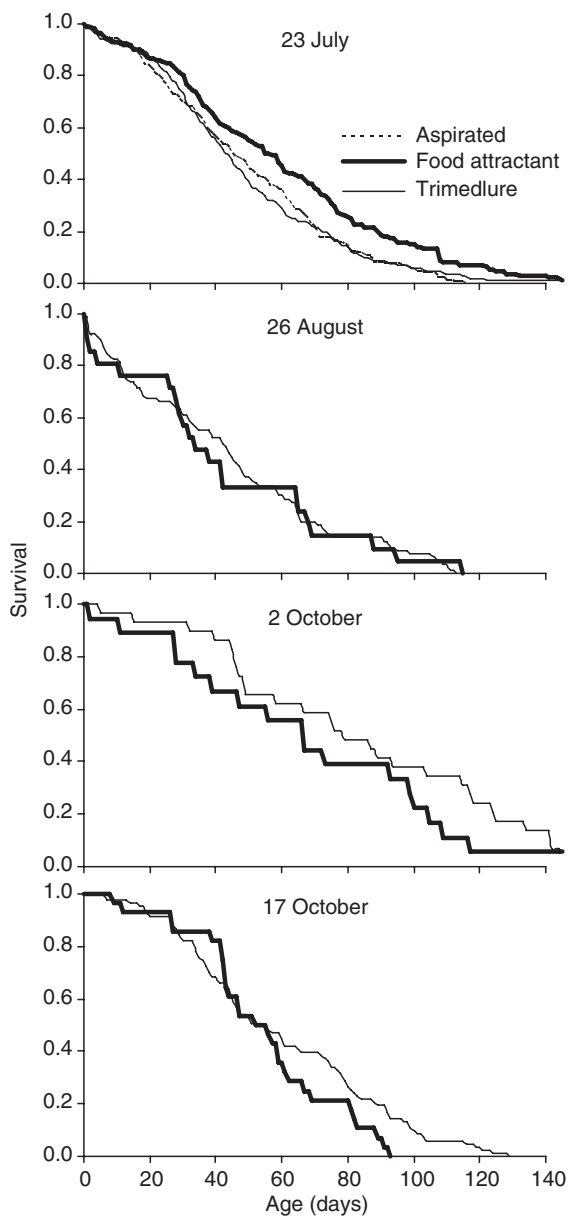


Figure 2 Post-capture survival of wild *Ceratitidis capitata* males collected on four dates with McPhail-type traps baited with a two-component synthetic food attractant, or a sex attractant. On one of these sampling dates, males were also collected with aspiration from host trees. Collections were made during the 2006 field season in a citrus orchard in Chios Island, Greece.

Age was not the only factor that influenced the degree of response to the traps in our experiments, also the feeding history of the flies played a big role. The overall response of adults that fed on sugar was nearly four-fold higher than that of flies fed on a protein diet and there was a strong interaction between type of food and all ages tested. The two-component lure in the trap is probably perceived as a

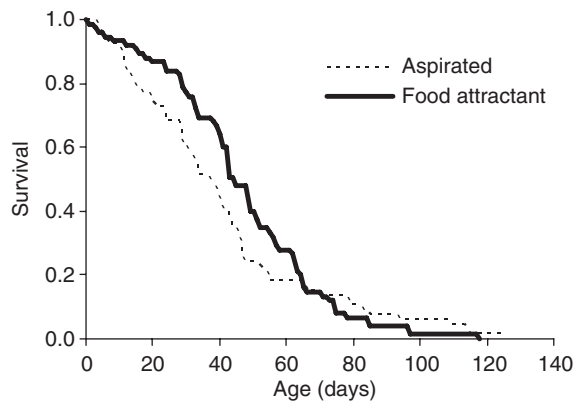


Figure 3 Post-capture survival of wild *Ceratitidis capitata* females collected on 23 July 2006 on Chios Island, Greece, using McPhail-type traps baited with a two-component synthetic food attractant or with aspiration from host trees.

cue for protein-rich food sources, such as bacteria and avian fecal material, which both sexes need (Bateman & Morton, 1981; Drew et al., 1983; Hendrichs et al., 1991). Response to this trap is evidently strongly affected by the nutritional state of adults, as this is defined by the quality and availability of food in an area. However, the motivation of foraging flies at a given time may also be controlled by other factors, such as sexual maturation, successful mating, social context, and an array of (other) physiological factors (Miller & Strickler, 1984; Browne, 1993). Identification of these factors would help better interpret capture data and thus make more accurate predictions about the age-structure of wild populations.

Our field experiment corroborated recent findings by Carey et al. (2008) that major changes in population age-structure occur in wild *C. capitata* populations on Chios Island, Greece. In that study, the availability of ovipositional hosts was the primary driver of populations. Monitoring population changes with a dependable trapping system is of great interest for the control of *C. capitata*. During sterile insect technique operations, high numbers of mass-reared sterile males are released in a wide area in high sterile to wild over-flooding ratios. Females that mate with sterile males lay unfertilized eggs that do not develop into larvae and thus the population declines. Evidence suggests that the sexual competitiveness of male *C. capitata* (N Papadopoulos, unpubl.) and other species targeted with this method (e.g., Abila et al., 2003) is significantly influenced by age. An adequate understanding of this influence could lead to increased efficiency of the method. If the age-structure of the target population is inferred with a sampling technique corrected for age bias, the over-flooding ratios could be adjusted to match the population of wild males that are in a reproductively active age.

In our experiment, aspirated flies were collected from a host tree while feeding in the late afternoon, which is the period of the day when most of the feeding activity of *C. capitata* has been documented to occur (Hendrichs et al., 1991). As adults require constant intake of food for maintenance, it was assumed that all age classes would forage on natural feeding arenas and that aspiration from a fruiting host tree would provide a sample comprising all ages. Such samples should theoretically be more representative than samples obtained by traps that exclude certain ages, such as traps that mainly lure sexually mature individuals. However, as evidenced by the differences in post-capture survival times, aspirated males did not differ from flies trapped with Trimedlure. Also, both aspirated males and females were relatively older at collection compared with flies captured with food-baited traps. The reason for this may be that young agile individuals escape capture by aspiration, compared with older individuals, and thus become underrepresented in a sample. Another possibility is that young flies may be less likely to forage for food on host trees compared with sexually mature flies. In addition to this putative exclusion of some proportion of young adults, aspiration has two other important disadvantages. First, it can only be used when populations are dense, because under low population flies are extremely difficult to locate visually. Second, aspiration is very demanding for time and personnel. In terms of cost effectiveness it cannot beat traps which employ powerful lures and capture large numbers of insects with minimal effort.

In temperate areas, detection of *C. capitata* under low population densities is hard even by trap systems regarded as highly effective (Carey, 1996; Papadopoulos et al., 2001). Our finding that food-baited McPhail traps capture younger flies compared with other methods suggests that these traps would be more effective for early detection of low spring populations, which mainly consist of young individuals originating from overwintering larvae. These traps would therefore be very valuable as aids for other management activities that work better when applied against incipient populations. In fact, in a study conducted in northern Greece, Papadopoulos et al. (2001) showed that Jackson traps baited with Trimedlure were by far outperformed by food-baited McPhail traps in early detection and total captures. On the other hand, the fact that in our field cage experiment older flies with access to protein tended not to respond as well suggests that the McPhail trap may be less effective for suppression purposes through mass trapping. In fact, in the study by Papadopoulos et al. (2001), after mid October the McPhail traps were less effective than the Jackson traps. By that time the population had grown considerably and was probably older.

Understanding age effects on capture techniques is of utmost importance for several categories of insects, medically important groups notwithstanding. For example, the longer a mosquito lives, the more likely she is to encounter an infectious host, survive the incubation period, and transmit a disease during subsequent feeding attempts (Dye, 1992). Because small changes in age-structure can result in large changes in transmission (Styer et al., 2006), accurate knowledge of age-structure is crucial for predicting the epidemiology of a disease. Our novel approach for estimating possible age-dependent bias in insect collection methods is based on life-table assay methods to estimate relative differences in the age compositions of captured flies. This straightforward and inexpensive assay can be adapted for any small arthropod species derived from the field with various sampling techniques. The association between age and sampling probability demonstrated in the present study is probably not specific to this particular system, but can be generalized from tephritids and *Mutillure* traps to other groups of insects of agricultural or medical importance that are commonly trapped for research and management purposes. More data from other systems will be needed to further assess the generality of our findings.

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