Effects of Food Deprivation on Attraction of Mexican Fruit Flies (Diptera: Tephritidae) to Grapefruit in a Wind Tunnel

DAVID C. ROBACKER[1] AND IVICH FRASER

Crop Quality and Fruit Insects Research, USDA–ARS, Kika de la Garza SARC, 2413 E. Highway 83, Building 200, Weslaco, TX 78596

Ann. Entomol. Soc. Am. 94(6): 954–958 (2001)

ABSTRACT Sexually mature, mated female Mexican fruit flies, *Anastrepha ludens* (Loew), from a laboratory culture, whether starved or not, were attracted to grapefruit in a wind tunnel. Females starved for 24 h were more strongly attracted than fed females but those starved for 48 h were equivalent to fed females in responsiveness. There were no significant differences in propensity to attempt oviposition on grapefruit by fed or starved females. Sexually mature, mated males, whether fed or starved for 24 h, were not attracted to grapefruit, but those starved for 48 h were attracted.

KEY WORDS *Anastrepha ludens*, fruit fly, grapefruit, attraction, oviposition, food deprivation

MEXICAN FRUIT FLY, *Anastrepha ludens* (Loew), is a highly polyphagous tephritid with ≈35 hosts that are infested in the field (Norrbom and Kim 1988). Among commercial citrus fruits, none of which are native hosts, grapefruit (*Citrus paradisi* MacFayden) is the preferred host of this fly in the sense that grapefruit orchards typically have much greater infestation rates than other types of citrus (Baker et al. 1944). This fact suggests that grapefruit should be highly attractive to gravid females foraging for oviposition hosts. Attraction of ovipositing females to host fruit odors has been reported for many species of fruit flies (Fletcher and Prokopy 1991, Jang and Light 1996).

In preliminary work to determine whether grapefruit is more attractive than other citrus fruits, we tested attractiveness of grapefruits in a wind tunnel to sexually mature, mated Mexican fruit flies fed ad libitum with sugar and yeast hydrolysate. We expected that flies fed this protein-rich diet would be gravid (Fletcher 1987, Tsitsipis 1989) and thus motivated to search for fruit to lay eggs as was suggested by Prokopy et al. (1998) for protein-fed Mediterranean fruit flies attracted to coffee fruit. However, attraction was weak in these bioassays.

Because grapefruit is a preferred host of this fly and because grapefruit was not very attractive to food-satiated flies in preliminary work, we hypothesized that sugar deprivation may enhance ovipositional attraction of gravid female flies to grapefruit. Previously, Robacker et al. (1990) reported that sugar deprivation enhanced attraction of Mexican fruit flies to the odor of fermented yellow chapote fruit, *Sargentia greggii* S. Wats., a native host of the fly, although oviposition responses of flies were not investigated.

Two experiments were conducted to test the hypothesis that sugar deprivation may enhance attraction of female Mexican fruit flies to grapefruit for oviposition. First, flies with different lengths of food-deprivation were tested for attraction to grapefruit in a wind tunnel. Second, the effect of food deprivation on propensity to attempt oviposition after arriving on a grapefruit was evaluated in small cages.

[1] E-mail: drobacker@weslaco.ars.usda.gov

Materials and Methods

Insects and Handling Methods. Laboratory stock of *A. ludens* was started in 1997 from 2,000 pupae collected from yellow chapote, a native host, from the Montemorelos area of Nuevo Leon in northeastern Mexico. Flies used in these experiments were reared on artificial medium and adults were held in 20.5 by 20.5 by 20.5 cm Plexiglas cages with screened tops containing a diet mixture of sugar and yeast hydrolysate, and with water supplied separately. Flies used in experiments were 10–20 d old. This is a reasonable age range for research on oviposition behavior because sexual maturation and mating occurs by 9 d posteclosion in laboratory-reared Mexican fruit flies (Dickens et al. 1982). Also, egg collection at our rearing facility is initiated when females reach 10 d posteclosion, although oviposition behavior is often observed in younger females. Laboratory conditions where flies were housed were 22 ± 2°C, and 50 ± 20% RH with a photophase of 0630 to 1930 hours provided by fluorescent lights. Experiments were conducted between 0900 and 1700 hours.

Wind-Tunnel Experiments. The purpose of these experiments was to determine the effect of food deprivation on attraction of Mexican fruit flies to grapefruit. Bioassays were conducted in a Plexiglas wind tunnel with the dimensions of 0.3 by 0.3 by 1.2 m. Each end of the wind tunnel was screened to allow airflow. The downwind end contained a baffle system to create a uniform airflow through the chamber. Air was pulled through the chamber at 0.4 m/s by an exhaust fan connected to the downwind end. The top of the chamber had two circular openings (12.8 cm diameter) with Plexiglas covers, located at each end of the chamber, to allow easy access to the chamber's interior. A 75-W "soft white" light bulb (General Electric, Cleveland, OH) in a reflecting lamp was positioned 17 cm above the downwind end of the chamber. The purpose of this light was to use the flies' positive phototactic reaction to minimize random flying into the upwind end of the chamber that could result in accidental landings on the fruit or the ball. Overhead lighting was provided by two banks of four fluorescent "cool white" lights each (F40CW, General Electric).

The grapefruits used as the ovipositional attractant source in wind-tunnel bioassays were variety Rio Red. Only ripe grapefruits of yellow color were used. They were picked from an orchard near the laboratory on the day before testing and were discarded each day after tests were done. Grapefruits were washed with water before each trial to remove any chemicals left by flies in the previous trial. A circular piece of the grapefruit rind and pulp measuring 2.5 cm in diameter was removed so that volatiles from both the peel and pulp were present in the aroma. A plastic yellow ball (Great Lakes Integrated Pest Management [IPM]), Vestaburg, MI) was used in place of grapefruit as a control in half the bioassays. Balls were 8 cm in diameter and were described previously (Robacker 1992).

In preparation for the experiments, either 24 or 48 h before testing, flies were transferred into pint-size, cylindrical paper cups, ≈12 flies of each sex per cup. One set of cups (controls) contained sugar and yeast hydrolysate and the other contained no food, for both the 24 and 48 h deprivation treatments. All cups were sprayed with water twice daily. Experiments were conducted as a series of four treatments tested in random order: fed flies offered grapefruit; fed flies offered a plastic yellow ball; starved flies offered grapefruit; and starved flies offered a plastic yellow ball. A grapefruit or a ball that served as a control was hung from the opening in the upwind end of the chamber, and one cup of flies per trial was placed under the downwind opening. Flies were allowed 5 min to leave the cup and respond to the fruit or ball. Upwind movement was scored if flies passed a point two-thirds of the distance from the release cup to the fruit or ball. Contact was scored for either landing or walking onto the fruit or ball. Attempted oviposition was scored for ovipositor probing whether or not penetration was achieved. No attempt was made to recover eggs. In experiment 1 we tested flies starved for 24 h against fed flies and in experiment 2 we tested flies starved for 48 h versus fed flies. Twenty-seven replications of the 24-h deprivation experiment and 28 of the 48 h deprivation experiment were conducted.

Small-Cage Experiment. The purpose of this experiment was to determine if starvation affected propensity of female Mexican fruit flies to attempt oviposition in grapefruit. At 24 h before testing, mated female flies were put into pint-size paper cups containing either sugar and yeast hydrolysate or no food. Cups were sprayed with water as in the previous experiments. To conduct the experiment, individual fed or starved flies were transferred into 30.5 by 30.5 by 30.5-cm aluminum-framed, aluminum-screened cages containing a whole (undamaged) ripe grapefruit (Rio Red). Fruit were washed with water before each bioassay. Oviposition behavior was observed for 1 h. Eighteen replications of each treatment were tested in pairs.

Statistical Analyses. Wind-tunnel experiments were analyzed by analysis of variance (ANOVA) using SuperANOVA (Abacus Concepts 1989). Proportions of flies that moved upwind, contacted the fruit or ball, or attempted oviposition, were transformed by arcsine of the square root (Snedecor and Cochran 1967) for statistical analyses. Proportions of 0 and 1 were replaced with 1/4n and (n - 1/4)/n, respectively, before transformation. The overall treatment effect (the four fruit type/deprivation treatments) was calculated then partitioned into fruit type effect, deprivation effect, and their interaction. Fruit type by time of day, deprivation state by time of day, and sex by time of day interactions were calculated in additional analyses of variance. Regressions of fly responses on fly age also were conducted in separate analyses.

The effects of food deprivation on the time elapsed until the first oviposition bout (latency), the time spent on grapefruit, and the time spent attempting to oviposit in the small-cage experiment were analyzed by *t*-tests. The percentage of females of each type to land on fruit at least once was analyzed by the chi-square test of significance of a binomial proportion (Snedecor and Cochran 1967).

Table 1. Effect of food deprivation on number of Mexican fruit flies that moved upwind, contacted a plastic yellow ball or a grapefruit, or attempted oviposition in a plastic yellow ball or a grapefruit, in a wind tunnel: females; 1-d deprivation (experiment 1)

	Moved upwind	Contacted source	Attempted oviposition	Attempted ovipositions per contact
Fed, yellow ball	3.1a	0.0a	0.0a	
Fed, grapefruit	7.6b	3.2b	2.2b	68.8a
Starved, yellow ball	3.1a	0.3a	0.3a	
Starved, grapefruit	13.8c	7.2c	3.1b	42.2a

Values in the first three columns are mean percentages responding out of the total females in the trial, and values in the last column are mean percentages attempting oviposition out of females that arrived on fruit. Means followed by different letters in the same column are significantly different at the 5% level by Fisher's protected LSD.

Results

Wind-Tunnel Experiments. Results for females in the wind-tunnel experiment testing effect of 24 h of food deprivation are shown in Table 1. With regard to effect of fruit type, more females, both fed and starved, moved upwind ($F = 24.9$; df = 1, 104; $P < 0.0001$), landed on the fruit ($F = 32.4$; df = 1, 104; $P < 0.0001$) and attempted to oviposit ($F = 13.7$; df = 1, 104; $P < 0.001$), in trials with a grapefruit than in trials with a yellow ball as a surrogate fruit. With regard to the

Table 2. Effect of food deprivation on number of Mexican fruit flies that moved upwind or contacted a plastic yellow ball or a grapefruit in a wind tunnel: males; 1-d deprivation (experiment 1)

	Moved upwind	Contacted source
Fed, yellow ball	3.2	0.3
Fed, grapefruit	5.1	0.0
Starved, yellow ball	2.2	0.3
Starved, grapefruit	4.7	0.8

Values are mean percentages responding out of the total males in the trial. No means were significantly different at the 5% level by Fisher's protected LSD.

Table 4. Effect of food deprivation on number of Mexican fruit flies that moved upwind or contacted a plastic yellow ball or a grapefruit in a wind tunnel: males; 2-d deprivation (experiment 2)

	Moved upwind	Contacted source
Fed, yellow ball	3.9a	0.0a
Fed, grapefruit	4.2a	0.6a
Starved, yellow ball	2.7a	0.9a
Starved, grapefruit	11.5b	6.4b

Values are mean percentages responding out of the total males in the trial. Means followed by different letters in the same column are significantly different at the 5% level by Fisher's protected LSD.

effect of 24 h of food deprivation on responses to grapefruit, more starved females moved upwind toward ($F = 11.2$; df = 3,104; $P < 0.0001$) and landed on grapefruit ($F = 13.7$; df = 3, 104; $P < 0.0001$), compared with fed females. There was no significant difference between the number of starved females and fed females that attempted to oviposit in the grapefruit. The number of starved females that attempted to oviposit after they were on grapefruit also was not significantly different from that of fed females ($F = 1.8$; df = 1, 21; $P = 0.20$). No more starved females than fed females moved upwind, or landed on or attempted to oviposit into the yellow ball.

Results for males in the wind-tunnel experiment testing the effect of 24 h of food deprivation are shown in Table 2. Males were not significantly attracted to grapefruit and food deprivation had no effect on their behavior.

Results for females in the wind-tunnel experiment testing the effect of 48 h of food deprivation are shown in Table 3. More females, both fed and starved, moved upwind ($F = 15.5$; df = 1, 108; $P < 0.0001$), landed on the fruit ($F = 24.1$; df = 1, 108; $P < 0.0001$) and attempted to oviposit in the fruit ($F = 9.4$; df = 1, 108; $P < 0.01$), in trials with grapefruit, than in trials with a yellow ball. No more starved females than fed females moved upwind, or landed on, or attempted to oviposit into either the yellow ball or grapefruit. The number of starved females that attempted to oviposit after they were on grapefruit also was not significantly different from that of fed females ($F = 1.6$; df = 1, 17; $P = 0.22$).

Results for males in the wind-tunnel experiment testing the effect of 48 h of food deprivation are shown in Table 4. More starved males moved upwind toward ($F = 5.1$; df = 3, 108; $P < 0.01$) and landed on ($F = 10.2$; df = 3, 108; $P < 0.0001$) grapefruit than yellow balls, and more starved males than fed males moved upwind toward and landed on grapefruit. No more starved males than fed males moved upwind or landed on the yellow ball.

No interactions involving time of day were significant. Regressions of fly responses on fly age also were not significant.

Small-Cage Experiment. Oviposition behavior on grapefruit by fed females and females deprived of food for 24 h is shown in Table 5. None of the measures of propensity of fed and starved females to spend time and attempt oviposition on grapefruit were significantly different.

Discussion

Behavior of flies in wind-tunnel bioassays varied both by food deprivation treatment and by sex. Females deprived of food for 24 h were more attracted than fed females to grapefruits (Table 1), but females deprived of food for 48 h were not more attracted than fed females (Table 3). These results suggest that the additional day of deprivation reversed the stimulatory effect of 24 h of deprivation. However, direct comparison of females starved for 24 and 48 h is needed to confirm this effect. With males, results were the opposite. Males deprived of food for 24 h did not respond

Table 3. Effect of food deprivation on number of Mexican fruit flies that moved upwind, contacted a plastic yellow ball or a grapefruit, or attempted oviposition in a plastic yellow ball or a grapefruit, in a wind tunnel: females; 2-d deprivation (experiment 2)

	Moved upwind	Contacted source	Attempted oviposition	Attempted ovipositions per contact
Fed, yellow ball	5.1a	0.0a	0.0a	
Fed, grapefruit	10.2b	4.0b	2.1b	53.3a
Starved, yellow ball	3.7a	0.0a	0.0a	
Starved, grapefruit	12.2b	4.8b	1.7ab	25.9a

Values in the first three columns are mean percentages responding out of the total females in the trial, and values in the last column are mean percentages attempting oviposition out of females that arrived on fruit. Means followed by different letters in the same column are significantly different at the 5% level by Fisher's protected LSD.

Table 5. Effect of 1 d of food deprivation on oviposition behavior of female Mexican fruit flies on grapefruit during 60-min tests in small cages

	Fed	Starved
Percentage to land on fruit	77.8	94.4
Percentage to attempt oviposition (among females on fruit)	100	100
Time until first oviposition bout (min) (among females that attempted oviposition)	14.5	4.9
Number of oviposition bouts	3.7	3.7
Total time attempting oviposition (min)	15.2	16.4
Total time on fruit (min)	42.7	45.4

No percentages or means are significantly different at the 5% level by the chi-square test of binomial proportions (percentages) or *t*-tests (means).

to grapefruit. However, after an additional day of food deprivation, males were significantly attracted to grapefruit. We hypothesize that different reasons for attraction to grapefruit account for differences in attraction responsiveness with changes in hunger. Both sexes feed on fruit (Aluja et al. 1989). In addition, females lay eggs whereas males sometimes search for ovipositing females with which they attempt forced matings (Robacker et al. 1991).

That food deprivation increases attraction of Mexican fruit flies to grapefruit is not surprising. Food deprivation has been shown to affect attraction to many types of hosts and foods of fruit flies (Liu and Chang 1995, Prokopy et al. 1996). However, here we present evidence that fed and starved females in close proximity to grapefruit exhibit nearly identical oviposition behavior (Table 5). Therefore, if food-deprived females are more attracted to the fruit, then it follows that more ovipositions may be made by hungry females than satiated ones, or at least by females that were hungry when they began their foraging bout. Although we have no data to support this contention, the data in Tables 1, 3, and 5 indicate that food deprivation does not diminish the number of oviposition attempts.

From an evolutionary perspective, why should females wait until they are hungry to search for oviposition resources? One possible answer is that it is a way of decreasing risk of predation. Any time a female leaves a hiding place to forage for fruit, she is placing herself in danger of predation. If females remain safely hidden until they become hungry, they can then combine feeding and oviposition bouts into a single foraging expedition, cutting their predation risk considerably. Alternatively, increased oviposition by hungry flies may be due to happenstance. Perhaps hungry females are simply more highly motivated than fed ones to search for fruit because carbohydrate deprivation quickly leads to death in fruit flies (Tsitsipis 1989). Increased oviposition would result because more starved females than fed ones are present on the fruit.

It is well documented that protein feeding increases fecundity of fruit flies (Fletcher 1987, Tsitsipis 1989). Therefore, it seems logical that protein feeding should increase attraction of female fruit flies to hosts for oviposition, as was discussed in the introduction. Our results seem contrary to this hypothesis. Actually, the hypothesis may be sound, but it may also be true that some hunger could increase overall oviposition rates by increasing attraction to, and thus amount of time spent on, host fruit. In fact, the likely explanation that ties the two ideas together is that the brief deprivation used in our work may increase sugar hunger to a much greater degree than protein hunger. Thus, the flies deprived of food for 24–48 h may be substantially sugar hungry but only very slightly hungry for protein. Previous results obtained in our laboratory are consistent with this idea. Typically, we need to deprive flies of protein from eclosion to see measurable behavioral effects in immature flies and for a week with mature flies, whereas 1–2 d of sugar deprivation results in strong foraging for sugar sources and longer deprivation quickly leads to death (Robacker 1991, 1998; Robacker et al. 1990; Robacker and Garcia 1993).

Fruit flies deprived of food are known to be easily stimulated to flight, apparently for the purpose of finding food (Prokopy and Roitberg 1984). However, that was not what we observed in this work. In both deprivation experiments, food deprivation did not increase the tendency of either males or females to initiate flights, as indicated by the equivalent upwind-movement responses of fed and hungry flies to yellow balls (Tables 1–4). In the Mexican fruit fly, it has been shown that flies starved for sugar or protein are more strongly attracted to fruit-type odors or bacterial odors, respectively (Robacker et al. 1990, Robacker and Garcia 1993, Robacker and Moreno 1995). Thus, we expected to observe an increase in attraction to grapefruit by flies deprived of food. The surprising finding was that, whereas females starved for 24 h were more responsive to grapefruit than fed females, those starved for 48 h were not. We first considered that 48 h of deprivation may have left the flies weak and not as able to fly as females starved for less time. However, males deprived of food for 48 h were much more responsive to grapefruit odor than either fed males or males starved for only 24 h (Tables 2 and 4). Obviously males and females were affected differently by food deprivation. We wondered if females were more weakened by deprivation than males so we analyzed the mortality of males and females held for either 24 or 48 h without food. After 24 h of deprivation, no mortality of either males or females occurred, and after 48 h, males suffered 29% and females 31% mortality. Thus, starvation affected survival of males and females equally in our experiments. This suggests that weakness may not be the answer, or at least not the whole answer.

Regardless of deprivation treatment, attraction of Mexican fruit flies to grapefruit was very weak. However, published reports on wind tunnel responses of various fruit flies to hosts are generally comparable, with percentage responding increasing with test duration (Averill et al. 1988, Jang and Light 1991, Landolt et al. 1992). Percentages of flies responding can be considerably higher when sex pheromone, parapheromones, or even synthetic food attractants are used as the odor stimuli (Landolt et al. 1992, Meats and Hartland 1999, Robacker 1999).

Although responsiveness of Mexican fruit flies to various types of attractants is known to vary with time of day (Robacker and Garcia 1990, 1993; Robacker et al. 1990; Robacker 1998), no time of day effect was proven in the current work. Fly age also had no effect on attraction to grapefruit in this study, probably because only sexually mature, mated flies between the ages 10–20 d posteclosion were used.

Acknowledgments

We thank Bob Heath (USDA-ARS, Miami, FL) and Peter Landolt (USDA-ARS, Wapato, Washington) for use of the

wind tunnel. Use of a product brand in this work does not constitute an endorsement by the USDA.

References Cited

Abacus Concepts. 1989. SuperANOVA. Abacus Concepts, Berkeley, California.

Aluja, M., M. Cabrera, J. Guillen, H. Celedonio, and F. Ayora. 1989. Behaviour of *Anastrepha ludens, A. obliqua* and *A. serpentina* (Diptera: Tephritidae) on a wild mango tree (*Mangifera indica*) harbouring three McPhail traps. Insect Sci. Applic. 10: 309–318.

Averill, A. L., W. H. Reissig, and W. L. Roelofs. 1988. Specificity of olfactory responses in the tephritid fruit fly, *Rhagoletis pomonella.* Entomol. Exp. Appl. 47: 211–222.

Baker, A. C., W. E. Stone, C. C. Plummer, and M. McPhail. 1944. A review of studies on the Mexican fruitfly and related Mexican species. USDA Misc. Publ. 531.

Dickens, J. C., E. Solis, and W. G. Hart. 1982. Sexual development and mating behavior of the Mexican fruit fly, *Anastrepha ludens* (Loew). Southwest. Entomol. 7: 9–15.

Fletcher, B. S. 1987. The biology of the Dacine fruit flies. Annu. Rev. Entomol. 32: 115–144.

Fletcher, B. S., and R. J. Prokopy. 1991. Host location and oviposition in tephritid fruit flies, pp. 139–171. *In* W. J. Bailey and J. Ridsdill-Smith [eds.], Reproductive behaviour of insects: individuals and populations. Chapman & Hall, New York.

Jang, E. B., and D. M. Light. 1991. Behavioral responses of female oriental fruit flies to the odor of papayas at three ripeness stages in a laboratory flight tunnel (Diptera: Tephritidae). J. Insect Behav. 4: 751–762.

Jang, E. B., and Light, D. M. 1996. Olfactory semiochemicals of tephritids, pp. 73–90. *In* B. A. MacPheron, and G. J. Steck [eds.], Fruit fly pests. St. Lucia Press, Delray Beach, FL.

Landolt, P. J., H. C. Reed, and R. R. Heath. 1992. Attraction of female papaya fruit fly (Diptera: Tephritidae) to male pheromone and host fruit. Environ. Entomol. 21: 1154–1159.

Liu, Y. C., and C. T. Chang. 1995. Attraction of food attractants to melon fly, *Dacus cucurbitae* Coquillett. Chin. J. Entomol. 15: 69–80.

Meats, A., and C. L. Hartland. 1999. Upwind anemotaxis in response to cue-lure by the Queensland fruit fly, *Bactrocera tryoni.* Physiol. Entomol. 24: 90–97.

Norrbom, A. L., and K. C. Kim. 1988. A list of the reported host plants of the species of *Anastrepha* (Diptera: Tephritidae). USDA-APHIS-PPQ APHIS 81–52.

Prokopy, R. J., and B. D. Roitberg. 1984. Foraging behavior of true fruit flies. Am. Sci. 72: 41–49.

Prokopy, R. J., X. Hu, E. B. Jang, R. I. Vargas, and J. D. Warthen. 1998. Attraction of mature *Ceratitis capitata* females to 2-heptanone, a component of coffee fruit odor. J. Chem. Ecol. 24: 1293–1304.

Prokopy, R. J., S. S. Resilva, and R. I. Vargas. 1996. Post-alighting behavior of *Ceratitis capitata* (Diptera: Tephritidae) on odor-baited traps. Fla. Entomol. 79: 422–428.

Robacker, D. C. 1991. Specific hunger in *Anastrepha ludens* (Diptera: Tephritidae): Effects on attractiveness of proteinaceous and fruit-derived lures. Environ. Entomol. 20: 1680–1686.

Robacker, D. C. 1992. Effects of shape and size of colored traps on attractiveness to irradiated, laboratory-strain Mexican fruit flies (Diptera: Tephritidae). Fla. Entomol. 75: 230–241.

Robacker, D. C. 1998. Effects of food deprivation, age, time of day, and gamma irradiation on attraction of Mexican fruit flies (Diptera: Tephritidae) to two synthetic lures in a wind tunnel. Environ. Entomol. 27: 1303–1309.

Robacker, D. C. 1999. Attraction of wild and laboratory-strain Mexican fruit flies (Diptera: Tephritidae) to two synthetic lures in a wind tunnel. Fla. Entomol. 82: 87–96.

Robacker, D. C., and J. A. Garcia. 1990. Responses of laboratory-strain Mexican fruit flies, *Anastrepha ludens*, to combinations of fermenting fruit odor and male-produced pheromone in laboratory bioassays. J. Chem. Ecol. 16: 2027–2038.

Robacker, D. C., and J. A. Garcia. 1993. Effects of age, time of day, feeding history, and gamma irradiation on attraction of Mexican fruit flies (Diptera: Tephritidae), to bacterial odor in laboratory experiments. Environ. Entomol. 22: 1367–1374.

Robacker, D. C., and D. S. Moreno. 1995. Protein feeding attenuates attraction of Mexican fruit flies (Diptera: Tephritidae) to volatile bacterial metabolites. Fla. Entomol. 78: 62–69.

Robacker, D. C., J. A. Garcia, and W. G. Hart. 1990. Attraction of a laboratory strain of *Anastrepha ludens* (Diptera: Tephritidae) to the odor of fermented chapote fruit and to pheromone in laboratory experiments. Environ. Entomol. 19: 403–408.

Robacker, D. C., R. L. Mangan, D. S. Moreno, and A. M. Tarshis Moreno. 1991. Mating behavior and male mating success in wild *Anastrepha ludens* (Diptera: Tephritidae) on a field-caged host tree. J. Insect Behav. 4: 471–487.

Snedecor, G. W., and W. G. Cochran. 1967. Statistical methods. The Iowa State University Press, Ames, IA.

Tsitsipis, J. A. 1989. Nutrition, pp. 103–109. *In* A. S. Robinson and G. Hooper [ed.], Fruit flies: their biology, natural enemies and control, vol.3A. Elsevier, New York.

Received for publication 8 January 2001; accepted 5 June 2001.