The biology of olive fruit fly, *Bactrocera oleae* (Rossi), was studied in the laboratory, greenhouse, and in canning olives, *Olea europaea* L., in relation to California regional climates. Adults survived in laboratory tests at constant temperatures and relative humidities of 5°C and 83%; 15°C and 59%; 25°C and 30%; and 35°C and 29% for 15, 6, 3, and 2 d without provisions of food and water and for 37, 63, 25, and 4 d with provisions, respectively. In a divided greenhouse, adults survived for 8–11 d in the warm side (36°C and 31% RH daytime); and in the cool side (26°C and 63% RH daytime) 10 d without provisions and 203 d with provisions. A significantly greater number of adults survived in the cool side than the warm side, and with provisions than without. First and last eggs were oviposited in olive fruit when females were 6 and 90 d old, respectively. The highest number of eggs was 55 per day in 10 olive fruit oviposited by 10 28 d-old females, with maximum egg production by 13–37 d-old females. A significantly greater number of ovipositional sites occurred in all sizes of immature green fruit when exposed to adults in cages for 5 d than 2 d. Adults emerged from fruit with a height of ≈1.0 cm or a volume of ≈0.2 cm³. More than seven adults per 15 fruit emerged from field infested fruit with a height of 1.1 cm and volume of 0.1 cm³. Larval length was significantly different among the first, second, and third instars and ranged from 0.7 to 1.6, 2.4–4.3, and 4.8–5.6 mm at 14°C; 0.8–1.1, 1.9–2.9, and 3.9–4.4 mm at 21°C, and 0.7–1.3, 2.4–2.9, and 4.4–4.8 mm at 26°C, respectively. Survival of pupae to the adult stage was significantly lower at 26°C than 14°C or 21°C. The period of adult emergence began at 38, 14, and 11 d over a period of 8, 5, and 1 d at 14, 21, and 26°C, respectively. Findings were related to the occurrence and control of California olive fruit fly infestations.

**KEY WORDS**  
*Bactrocera oleae*, reproductive biology, adult survival, temperature, humidity
ney Agricultural Research and Extension Center in Parlier, CA, in cooperation with the California Olive Committee, Fresno, CA, and from UC Davis, CA. Specific biological information was needed by the canned olive industry to help California growers manage the pest (Rice 2000). The biology of olive fruit fly in the United States could possibly differ from its Eastern Mediterranean subpopulation as it may have adapted to a new world region, in the same manner in which the three Mediterranean subpopulations diverged.

Daane and Johnson (2010) reviewed the olive fruit fly control literature and included studies of the pest in California. The distribution of olive fruit fly in different olive production regions of the state has been delineated with trapping programs (Burrack et al. 2011, Yokoyama et al. 2006) and different trapping techniques (Burrack et al. 2008). In general, high populations of olive fruit fly occur in cool, humid coastal areas, and low numbers of the pest are found in the hot, arid areas of California, such as the San Joaquin Valley where olives are grown for canning.

Upon detection of olive fruit fly, quarantine strategies were developed to control the larvae in harvested fruit transported to canning plants in northern California (Yokoyama and Miller 2004). After the pest was considered established, integrated pest management (IPM) programs were initiated in interior valleys (Johnson et al. 2006). The primary control tactic for olive fruit fly uses bait sprays with spinosad as the active ingredient, but Kakani et al. (2010) have reported the development of resistance to spinosad in some regions of California.

Resident natural enemies in California do not adequately control olive fruit fly populations (Daane 2011). A greater understanding of the life history of the pest in different regions of the state would assist with the search, match, and selection of imported natural enemies for biological control. An imported parasitoid, Psyttalia concolor (Silvestri), has been released in several regions and seasonal climates with rates of parasitism related to temperatures and host abundance (Yokoyama et al. 2008, 2010, 2011; Wang et al. 2011a). Wang et al. (2011b) related the developmental times between olive fruit fly and two parasitoids under different temperatures.

The distribution of olive fruit fly in California is primarily limited by the adverse effects of summer heat. Wang et al. (2009a, b) showed temperature regimes of 18.3°C and 35.0 or 37.8°C reduced egg production and lowered adult flight performance affecting abundance and dispersal. However, Johnson et al. (2011) found that olive fruit fly adults were able to overcome heat stress if provided with water and carbohydrates. Based on the adverse effects of hot weather, Gutierrez et al. (2009) predicted that the distribution of olive fruit fly would increase northerly and decrease southerly because of climate warming. Genç and Nation (2008) reported the optimum development of olive fruit fly immature stages from laboratory diet was 27°C. Information from their eastern Mediterranean population is useful in elucidation of the life history of olive fruit fly under different conditions in California.

In studies of host and insect interactions, Burrack and Zalom (2008) showed cultivar affected olive fruit fly oviposition and immature development and the large fruit of Sevillano, Manzanillo, and Mission cultivars were heavily infested. Within the fruit, Burrack et al. (2009) found that high egg and larval densities negatively impacted resultant pupal numbers and size as well as adult emergence. Although large fruit size supported infestation, the size of the infestation limited olive fruit fly immature development. Furthermore, fruit size was shown by Wang et al. (2009c, d) to affect parasitism by Psyttalia concolor (Szépligeti) and P. lounsburyi (Silvestri) because the large size of domesticated fruit was favorable for larval infestation and protected larvae from parasitoids with short ovipositors.

To provide canned olive growers with fundamental information that would help predict and evaluate life stages in olive orchards, factors were studied that would affect olive fruit fly development under conditions relevant to California. Physical conditions including temperature, and biological considerations such as the availability of food and water, that would affect olive fruit fly adult survival in the field were studied in the laboratory and greenhouse. To help growers determine fruit susceptibility to attack, the ovipositional period for olive fruit fly females and the acceptability of different sizes of olive fruit for oviposition and larval development were determined in the laboratory. Pupal survival at different temperatures was studied to evaluate the capacity of this life stage to overwinter and produce the next seasonal generation of adults. Larval length was selected as a practical method for growers to determine immature stages in fruit infestations, such as developed by Frick et al. (1954) for cherry fruit fly, B. indifferens Curran. Tests were performed in California Mission olives, a U.S. cultivar introduced from Mexico in 1769 (Sutter 1994), to represent olive fruit fly response in canned olives.

**Materials and Methods**

**Source of Insects.** Mission or Manzanillo olives that were infested with olive fruit fly larvae were collected from olive trees found in San Jose, Hollister, Arroyo Grande, Sylmar, and Los Angeles, CA. The infested fruit was placed in plastic containers (22 cm wide × 32 cm long × 13 cm high) and covered with organy fabric until the third instars emerged and pupated. The pupae were collected, placed in petri dishes, and held in the laboratory at ~25°C and 50% RH until adult emergence. Newly emerged adults were used in tests or placed in cages to infest olive fruit to obtain eggs and larvae when Mission fruit was available from September through January in different regions of California. Laboratory temperature and humidity were monitored with a hygrothermograph (model GT485, Omega Engineering, Stamford, CT).
Adult Longevity in Laboratory Tests. Four environmental chambers (model E32560, Lab-Line, Melrose Park, IL) with single temperature controls were set at 5, 15, 25, or 35°C with relative humidities set at 85, 65, 25, and 35%, respectively. Newly emerged olive fruit fly adults were placed in a cylindrical cardboard carton (477 ml) (No. HD16, Solo Cup, Urbana, IL) with the opening covered with organically fabric that was held in place with the collar of the lid. The carton was placed on its side and a plastic lid (8 cm diameter) was glued to the bottom of the cylinder as a pedestal.

Twenty-six, 1-d-old adults having a 1:1 sex ratio as determined by Moore (1962) were placed in each carton cage. The adults were provided with either no food or food in droplets of protein hydrolysate (4 g), mixed with sucrose (7 g), and water (100 ml) (Yokoyama et al. 1992), placed on the fabric cover. Each cage was considered a replicate and five replicates were used for each food treatment and temperature. Adults were evaluated daily or occasionally after 3 d (because of logistics) for survival in each cage until all adults died. The results were reported as mean ± SEM day adults survived among the replicates and compared between food treatments at each temperature with a t-test (GraphPad Software 2007). The chamber digital display of actual temperature and humidity were recorded at each observation and reported as the daily mean ± SEM.

Adult Longevity in Greenhouse Tests. Temperature and relative humidity were maintained in a partitioned glass greenhouse (5.6 m wide × 9.3 m long × 4.1 m high) as described by Yokoyama and Miller (2007), and monitored with a hygrothermograph. Temperatures and humidity were manipulated with exterior and interior shade cloths, swamp coolers, ground and overhead water misters, and propane and electrical heaters. The north side of the greenhouse was maintained at cool temperatures and high humidities throughout the day similar to summer, diurnal coastal conditions. Temperatures and humidities in the south side were maintained warm throughout the day similar to diurnal inland valley conditions. The daily maximum and minimum diurnal and nocturnal temperatures and relative humidities were reported as the mean (±SEM).

Temperatures and humidities in the greenhouse were compared with mean ± SEM high and low values calculated from California Irrigation Management Information System (CIMIS) weather data for July 2001–2010 for the Los Angeles Basin Station 174 and the San Joaquin Valley Station 80 selected to represent coastal and inland valley climates, respectively.

Polyethylene screen cages (35 cm wide × 35 cm long × 56 cm high) described by Yokoyama and Miller (2007) with an access opening (12 cm wide × 12 cm high) covered with a flap (30 cm wide × 30 cm high) were suspended inside a polyvinyl chloride pipe frame (2.7 cm diameter, 42 cm wide × 42 cm long × 65 cm high).

Thirty-five newly emerged olive fruit fly adults (1:1 sex ratio) were placed in each cage. The adults were provided with combinations of food in droplets of protein hydrolysate and a 5% sucrose solution of water in a vial at the top of the cage. Survival was tested on the cool side of the greenhouse in cages provided with either food and water (two replicates of six cages), or no food and no water (four replicates of 4–6 cages, 22 cages total); and, on the warm side of the greenhouse with either food and water (three replicates of six cages), or no food and no water (four replicates of six cages). Replicate tests were started at different dates. Survival in each cage was observed daily or occasionally after three or 4 d until all adults had died. The results were reported as percentage survival or the mean ± SEM percentage survival of 2–4 replicates per date (GraphPad Software 2007). Adult survival between the cool and warm side of the greenhouse, and with and without food and water were compared by survival analysis log-rank test (SAS Institute 2010).

Ovipositional Period. Ten female and 10 male newly emerged olive fruit fly adults were placed in a screened cage (30.5 cm wide × 30.5 cm long × 30.5 cm high). Ten mature, green Mission olives that were freshly picked in September through early November or refrigerated at ≈4.4°C for not >4 d were placed in each cage with the adults. The cages were held in the laboratory at 23°C and 48% RH relative humidity and monitored with a hygrothermograph. Fruit that was exposed to adults was removed from the cage and replaced with unexposed olives daily or occasionally after 3 or 4 d. Five cages were prepared in this manner and each cage was considered a replicate. The fruit that was exposed to oviposition was inspected for ovipositional sites, and each site was dissected and inspected for eggs. The total number of eggs oviposited by 10 females per cage were recorded daily or occasionally after 3 or 4 d and reported as the mean ± SEM of 2–5 replicates of 10 fruit, 0–90 d after adult emergence.

Development in Immature Fruit in the Laboratory. Immature green fruit was collected in 2–23 September from Mission olive trees in Sanger (36° 45′W; 119° 33′N), CA, and arbitrarily sorted into six visual size groups. For convenience, the groups were subjectively categorized as extra-small, small, medium, medium-large, and large. Sixty fruit of the smallest size and 30–35 fruit in the remaining five size groups were placed in separate screened cages (30.5 cm wide × 30.5 cm long × 30.5 cm high) in the laboratory, and 35 olive fruit fly adults (16–18 females) that were 3–21 d old at the start of each test were placed in each cage. A cage was considered a replicate and three replicates of each size group were exposed to adults for either 2 or 5 d. Temperature (≈23°C) and relative humidity were monitored with a hygrothermograph.

Three replicates of 15 fruit per cage were measured between the stem and blossom end and across the width to determine height and diameter, respectively. The estimated volume of each olive fruit was calculated using the formula for the volume of an elliptical spheroid, where volume = one-sixths times height times diameter² (Mutschler et al. 1986).

After exposure to olive fruit fly adults, the fruit from each cage was removed, inspected for ovipositional
sites, and placed on top of rigid plastic mesh inside a plastic container (15 cm wide × 15 cm long × 5 cm deep), and the top covered with organdy fabric held in place with the lid band. The olives were held for 170 d until all pupae and adults had emerged.

The number of ovipositional sites and adults in all size groups of olive fruit were compared after 2 and 5 d exposures with a two-tailed paired t-test, and fruit height and volume were compared among the size groups using a one-way analysis of variance (ANOVA) and Tukey’s test (GraphPad Software 2007). Fruit height, diameter, and volume, ovipositional sites, and number of adults in each of the six size groups of immature fruit were reported as the mean ± SEM of three replicates.

Development in Immature Fruit in the Field. Immature green fruit was collected on 14 September from Mission olive trees infested with olive fruit fly east of the Los Angeles International Airport (33° 56' N; 118° 22' W), CA. The height and diameter of 15 of the smallest green fruit were measured. The fruit was placed on plastic mesh inside a plastic container covered with organdy fabric. The olives were held in the laboratory at approximately 23°C and 50% RH for 170 d. Fruit height, diameter, and volume, ovipositional sites, and olive fruit fly adults emerging from the fruit were reported as the mean ± SEM of three replicates.

Development of Eggs and Larvae in Olives. Duration of olive fruit fly first through third instars in olive fruit was determined in environmental chambers. Sixty Mission olive fruit were placed in a cage for 24 h with olive fruit fly adults of variable age on a daily basis. The fruit was removed from the cage, and placed in plastic containers inside an environmental chamber set at one of three temperature and relative humidity combinations of 15°C and 65%; 21°C and 60%; and 25°C and 35%. Five daily determinations of temperature and relative humidity were recorded for the chamber set at 21°C and 60%. Temperature loggers (model XT10S-5 + 37, Onset Computer Corp., Bourne, MA) with external thermistors (model TMC6-1T, Onset Computer Corp.) on extension cables (1.8 m long), and humidity loggers (model SRHA08, Onset Computer Corp.) were placed inside chambers set at 15°C and 65%, and 25°C and 35%, and programmed to record 720 determinations per day. Daily temperature and relative humidity were reported as the mean ± SEM per day.

A plastic container of 60 olive fruit that had been previously exposed 1–3 d earlier to oviposition by olive fruit fly adults was removed from one of the three chambers. The fruit was inspected for ovipositional sites and dissected for eggs, and first through third instars. The length of each larval stage was recorded and the fruit in each container was inspected and dissected until at least 40 fruit were found with immature stages. Larval lengths were compared with separate instars at each temperature and relative humidity combination using a one-way ANOVA and Tukey’s test (GraphPad Software 2007).

Pupal Survival in Laboratory Tests. Newly emerged to 1 d-old pupae were placed in a plastic container. Each container was considered a replicate. Four replicates were tested in an environmental chamber at one temperature and relative humidity combination of 15°C and 65%; 21°C and 60%; and 25°C and 35% as described above with 120-200, 198-205, and 197-200 pupae per replicate, respectively. The containers were examined daily or occasionally after 3 d until all adults had emerged. Percentage survival was reported as the mean ± SEM of the replicates and compared between each temperature and humidity combination with an unpaired two-tailed t-test (GraphPad Software 2007).

Results

Adult Longevity in Laboratory Tests. Mean ± SEM temperatures in each environmental chamber were 5.0 ± 0, 15.0 ± 0, 25.0 ± 0, and 35.0 ± 0°C, with respective relative humidities of 83.1 ± 0.4, 59.3 ± 0.6, 29.7 ± 0.7, and 29.0 ± 1.6%. Among temperature and humidities tested, olive fruit fly adults lived the longest at 15°C without food and water (2 wks) at 5°C and 83% RH and with food and water (9 wks) at 15°C and 59% RH (Table 1). At 25°C and 30% RH, adults lived for 3 d without and >3 wks with food and water. Survival was lowest at 35°C and 29% RH; and adults lived for ≈2 d without and 4 d with food and water.

Adult Longevity in Greenhouse Tests. The daily mean ± SEM temperature and relative humidity in the warm side ranged from 36.2 ± 0.3°C and 31.4 ± 1.4% during the day, and 25.6 ± 0.3°C and 47.5 ± 2.8% at night; and in the cool side of the greenhouse ranged from 26.5 ± 0.2°C and 62.7 ± 0.9% during the day and 24.7 ± 0.2°C and 58.5 ± 1.5% at night. Lowest temperatures occurred at night and were similar on both sides of the greenhouse.

The range of maximum and minimum mean ± SEM temperatures and humidities based on CIMIS data in the center of the San Joaquin Valley was 35.8 ± 0.3–18.1 ± 0.3°C and 22.0 ± 0.6–70.3 ± 0.9% RH; and in the Los Angeles Basin was 26.3 ± 0.7–16.5 ± 0.5°C and 58.6 ± 1.8–92.5 ± 1.0% RH.

Olive fruit fly adults lived significantly longer (203 d) on the cool side of the greenhouse with food and water (Fig. 1) than without (χ² = 220.90; df = 1; P < 0.0001) (Fig. 2), and significantly longer than the warm side with (χ² = 195.76; df = 1; P < 0.0001) and without food and water (χ² = 177.34; df = 1; P <

<table>
<thead>
<tr>
<th>Temp. (°C)</th>
<th>% relative humidity</th>
<th>No solution*</th>
<th>Solutionb</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>83</td>
<td>14.6 ± 1.0</td>
<td>37.4 ± 1.59*</td>
</tr>
<tr>
<td>15</td>
<td>59</td>
<td>5.8 ± 0.2</td>
<td>62.8 ± 5.0*</td>
</tr>
<tr>
<td>25</td>
<td>30</td>
<td>3.4 ± 0.4</td>
<td>25.2 ± 3.5*</td>
</tr>
<tr>
<td>35</td>
<td>29</td>
<td>2.0 ± 0.0</td>
<td>4.2 ± 0.5*</td>
</tr>
</tbody>
</table>

* Significantly different than survival without solution (P < 0.05, t-test; GraphPad Software 2007).

b Four replicates of 23–29 adults.
Survival (8–11 d) was not significantly different between the cool side without food and water than the warm side with (\( \chi^2 = 0.05; df = 1; P = 0.8237 \)) and without food and water (\( \chi^2 = 0.01; df = 1; P = 0.9095 \)). On the warm side, survival was not significantly different (\( \chi^2 = 0.03; df = 1; P = 0.8687 \)) between adults provided with or without food and water.

**Ovipositional Period.** The first and last eggs were oviposited in olive fruit when olive fruit fly females were 6 and 90 d-old, respectively (Fig. 4). The highest mean ± SEM number of eggs per day in 10 olive fruit was 55 ± 24 and were oviposited by 10 28 d-old females. In all replicates, peak oviposition by females was observed between 13 and 37 d of age.

**Development in Immature Fruit in the Laboratory.** Mean ± SEM temperature and relative humidity in the laboratory was 22.7 ± 0.1°C and 47.9 ± 0.8%. Olive fruit height from the stem to the blossom end was significantly different (\( F = 272.0; df = 11, 24; P < 0.0001 \)) among five fruit size groups, and the calculated fruit volume was significantly different (\( F = 65.45; df = 11, 24; P < 0.0001 \)) among three of six fruit size groups (Table 2). A significantly greater number of olive fruit fly ovipositional sites (\( t = 3.21, df = 34, P = 0.0001 \)) was found in all size groups of immature green fruit when exposed to olive fruit fly adults in cages for 5 d than for 2 d. The number of F\(_1\) adults that emerged from the fruit was similar for both exposure periods. F\(_1\) adults emerged from fruit that had a height of ≥1.0 cm or a volume of ≥0.2 cm\(^3\). More adults (0.5 per 15 fruit) were reared from the largest size fruit (2.4 cm height) after a 5 d exposure than any other size group or exposure period.

**Development in Immature Fruit in the Field.** More than seven olive fruit fly adults per 15 fruit emerged from olive fruit with a height of 1.1 cm and volume of 0.1 cm\(^3\) (Table 2).

**Development of Eggs and Larvae in Olives.** Mean ± SEM daily temperatures and relative humidities in
showed that 2 h exposures to temperatures above 36°C adversely affected survival of olives grown for canning by infestations in olives grown for canning in commercial olive groves in the hot and arid California inland valleys (Johnson et al. 2011, Yokoyama et al. 2006, Wang et al. 2009a). However, environmental chambers were 14.07 ± 0.01°C and 66.17 ± 0.12%; 21.02 ± 0.04°C and 58.8 ± 1.07%; and 25.75 ± 0.01°C and 41.09 ± 0.06%.

Larval length was significantly different among the first through third instars at 14°C (F = 202.7; df = 2, 26; P < 0.0001), 21°C (F = 63.98; df = 2, 9; P < 0.0001), and 26°C (F = 234.4; df = 2, 10; P < 0.0001) (Fig. 5). Mean larval length of the first, second, and third instars ranged from 0.7 to 1.6, 2.4–4.3, and 4.8–5.6 mm at 14°C; 0.8–1.1, 1.9–2.9, and 3.9–4.4 mm at 21°C, and 0.7–1.3, 2.4–2.9, and 4.4–4.8 mm at 26°C, respectively.

Pupal Survival in Laboratory Tests. Survival of olive fruit fly pupae to the adult stage was significantly lower at 26°C and 41% than at 14°C and 66% (t = 4.881; df = 6; P = 0.0028), and at 21°C and 59% RH (t = 5.596; df = 6; P = 0.0014) (Table 3). At 14, 21, and 26°C, the period of adult emergence began at 38, 14, and 11 d over a period of 8, 5, and 1 d, respectively.

Discussion

This study provides basic information for evaluating olive fruit fly infestations in olives grown for canning in different regions of California. Olive fruit fly requires mild seasonal temperatures and a source of food and water for prolonged survival in incubator (Tables 1 and 3) and greenhouse tests (Figs. 1 and 2), while survival was adversely affected by hot and dry conditions (Fig. 3). These findings are consistent with Wang et al. (2009b) showing high temperature caused a loss of flight performance, especially in the absence of food and water, and a high temperature regime inhibited egg hatch (Wang et al. 2009a). Even brief exposures to high temperature can cause adverse effects. Recent work by Pappas et al. (2011) in Greece showed that 2 h exposures to temperatures above 36°C in the laboratory adversely affected survival and reproduction. In Portugal, Gonçalves and Torres (2010) discussed the adverse effects of high temperatures and low humidity and other physical and biological factors, such as fruit quality and maturation, on the use of degree-day models to predict olive fruit fly activity. Contemporary observations by Mediterranean workers help explain the very low numbers of olive fruit fly found in commercial olive groves in the hot and arid California inland valleys (Johnson et al. 2011, Yokoyama et al. 2006, Wang et al. 2009a).

Table 2. Mean ± SEM fruit dimensions, olive fruit fly ovipositional sites, and adults and percentage females that emerged from different sizes of immature fruit infested in cages for 2 or 5 d, or collected from olive trees in Los Angeles

<table>
<thead>
<tr>
<th>Exposure (d)</th>
<th>Height (cm)</th>
<th>Diameter (cm)</th>
<th>Vol. (cm³)</th>
<th>No. egg sites</th>
<th>No. adults per 15 fruit</th>
<th>% females</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>0.54 ± 0.01ab</td>
<td>0.75 ± 0.02</td>
<td>0.05 ± 0.00a</td>
<td>0.05 ± 0.01</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>0.56 ± 0.03bc</td>
<td>0.70 ± 0.03</td>
<td>0.05 ± 0.01a</td>
<td>0.13 ± 0.13</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Field</td>
<td>1.127 ± 0.004</td>
<td>0.876 ± 0.004</td>
<td>0.144 ± 0.002</td>
<td>—</td>
<td>7.2 ± 0.6</td>
<td>41.3 ± 1.8</td>
</tr>
</tbody>
</table>

Means within columns followed by different letters are significantly different (P < 0.05, Tukey’s test; GraphPad Software 2007).
predicting infestations by summer weather in California may be limited the numerous variables described by Gonçalves and Torres (2010).

Cool coastal conditions similar to the Los Angeles Basin or warm inland, San Joaquin Valley conditions were simulated in greenhouse temperatures and humidity regimes. Although the daytime temperatures in the greenhouse were similar to weather records in these regions, the night temperatures were not as low. The difference was caused by the methods used to artificially manipulate the greenhouse climate. On the warm side of the greenhouse, a mean temperature and humidity of 36°C and 31% during the day (Fig. 3) was similar to the highest temperature and humidity tested in environmental chambers (Table 1). Longevity of olive fruit fly adults ranged from 8 to 10 d, whether or not food and water was provided in the greenhouse tests, and only 2–4 d at constant temperatures in the laboratory study. The more natural conditions in the greenhouse including lighting and the lower temperatures and higher humidity at night may have mitigated the adverse effects of day time heat stress on adult survival.

In November 2008, a very large population of olive fruit fly was discovered in Lodi in the inland San Joaquin Valley. Thirteen adults were trapped per day and four larvae were recovered per olive fruit in this location (Yokoyama et al. 2010). In the summer month of July 2008, Lodi had three consecutive days of over 37.5°C high temperature (Station 166, Lodi West, CA Irrigation Management Information System) similar to the temperatures tested by Wang et al. (2009a), that could have caused high adult mortality because of heat stress (Johnson et al. 2011). However, the evening temperatures on these hot days averaged 18.5°C that probably relieved the detrimental effect of diurnal high temperatures on survival. Therefore, high temperatures alone may not limit the abundance of the pest. Wang et al. (2009a) showed that the adverse effects of heat can be offset by various behavioral adaptations that enhance survival in laboratory tests using a 3-d, 18.3–37.8°C high temperature regime. In addition, Yokoyama et al. (2012) found major olive fruit fly infestations in the San Joaquin Valley in cooler foothill locations and areas cooled by the marine influence of the Sacramento Delta.

On the cool side of the greenhouse, adults lived as long as 202 d when food and water were provided (Fig. 1). Under conditions of mild temperatures and high humidity olive fruit fly adults could survive for almost 7 mo if food and water were available. Based on these findings, very large populations could develop during the fall months when olive fruit is abundant, weather conditions are optimal, and the adults are long lived. Growers may need to implement control procedures to prevent crop damage.

The relationship between physical conditions and survival of olive fruit fly may help evaluate the capacity of other Bactrocera spp. to invade California. For example, the peach fruit fly, B. zonata, is an extremely destructive pest with multiple hosts, many of which are commercially grown in the California San Joaquin Valley. The physical requirements of peach fruit fly differ from olive fruit fly because the optimum temperatures for development are 25–30°C (OEPP/EPPO 2005) and 14–26°C (Tables 1 and 3; Figs. 1 and 5), respectively. Based on the data, we would anticipate olive fruit fly infestations in regions and seasons of cooler temperatures and peach fruit fly to prevail in warm weather conditions such as found in the interior valleys of California during the summer. In fact, the pest was detected and eradicated in June 2006 in Fresno in the San Joaquin Valley, the agricultural center of the state (USDA, APHIS 2006). The hot and dry California Central Valley was previously identified by Carey and Dowell (1989) as inhabitable by this exotic pest. Basic research describing the effect of temperature on the life history of fruit fly species such as olive fruit fly and peach fruit fly provides a valuable tool in identification of agricultural regions that may be vulnerable to invasions.

Food and water was a requirement for olive fruit fly adult longevity. Water may be a limited resource in the arid inland valley than in the humid coastal regions. A source of food for olive fruit fly adults in the field is honeydew produced by black scale, Saissetia oleae (Olivier), which is commonly found throughout the Central Valley of California (Daane and Caltagirone 1989). Johnson et al. (2011) found that black scale honeydew as a carbohydrate source helped olive fruit fly adults survive periods of hot weather.

Cold temperature is not a critical factor for olive fruit fly survival in the Mediterranean climate of California. Winter conditions in olive growing regions rarely drop below the critical low temperature of minus 6.5°C for olive fruit fly survival (Koveos 2001). Seasonal cold conditions would not necessarily limit pest distribution except when freezes cause fruit to drop from the trees eliminating larval habitats (Yokoyama et al. 2011).

The preovipositional period for olive fruit fly females was 6 d in this study (Fig. 4). Thereafter, peak egg production occurred when the females were 2–5 wks-old and females produced eggs for about 3 mo. This work shows that olive fruit fly can produce a very large number of eggs for a very long period and demonstrates the potential of the pest to cause severe infestations in olives over multiple generations in one season under favorable weather conditions. Control tactics should be implemented in commercial olive orchards before large adult populations develop.

### Table 3. Period of olive fruit fly adult emergence and percentage survival of pupae to adults when exposed to three constant temperatures and humidities in the laboratory

<table>
<thead>
<tr>
<th>Temp. (°C)</th>
<th>% relative humidity</th>
<th>Period of emergence, d</th>
<th>% survival (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>14</td>
<td>66</td>
<td>35–46</td>
<td>66.3 ± 9.1</td>
</tr>
<tr>
<td>21</td>
<td>59</td>
<td>14–19</td>
<td>64.2 ± 7.2</td>
</tr>
<tr>
<td>26</td>
<td>41</td>
<td>11–12</td>
<td>16.3 ± 4.6*</td>
</tr>
</tbody>
</table>

* Significantly different than survival at 15 and 21°C (P < 0.05, t-test; GraphPad Software 2007).
Calculation of fruit volume (Yokoyama et al. 2006) and determination of fruit pit hardening were proposed as methods to determine fruit susceptibility for initiation of bait sprays (Rice et al. 2003). Measuring fruit height is an easy method for growers to determine potential susceptibility to olive fruit fly infestation. Separation of immature fruit sizes was more readily done using fruit height from the stem to the blossom end than calculating fruit volume (Table 2). Olive fruit fly adults were found to develop from fruit that had a height of ≥1.0 cm in laboratory tests and 1.1 cm in field collected fruit. In olive orchards, olive fruit fly could possibly develop in smaller fruit than used in this study. Nonharvested fruit continues to grow increasing the availability of larval food. The fruit in this study were harvested simultaneously from the same trees which resulted in differences in fruit shapes. The quality of small fruit may also differ among cultivars. Nonetheless, using the criteria of successful development of oviposited eggs to the adult stage in harvested fruit, we found that olive fruit fly can complete development in small immature fruit and early control practices may be considered in areas with previous heavy infestations.

The largest immature fruit (2.4 cm height, 1.6 cm$^3$) produced more olive fruit fly adults, almost one per two olive fruit, than any other size tested at the longest exposure period (5 d) in laboratory tests (Table 2). When comparing olive cultivars, Burrack and Zalom (2008) reported that larger size olives may have a longer period and more volume for infestation than smaller cultivars. Mature fruit of large cultivars would support multiple olive fruit fly immatures, but the size of the fruit may limit biological control, especially by parasitoids with short ovipositors (Wang et al. 2009c, d).

Larval length can be easily used in the field to determine immature life stages that may be found in fruit and would help growers predict adult emergence and time control tactics. Although larval density may affect size (Burrack et al. 2009), the effect should be proportionate among instars in the same fruit. Larval food may also affect size and we observed wide variation in individual length within the instars during growth. Others have used morphological methods to separate the instars including the presence or absence of prothoracic spiracles (Neuenschwander et al. 1986) and by the shape and color of the mandibular stylets (Wang et al. 2009d). However, such methods may be impractical for use by growers.

Laboratory studies of olive fruit fly eggs and larvae showed slow development at 14°C (Fig. 5) which is similar to the finding of Genc and Nation (2008) who reported the slowest development of immatures for their eastern Mediterranean population was at 16°C in laboratory diet. Although development of the immature stages required >1 mo in the fruit at 14°C, the adults lived longer when provided with a food and water solution at 15°C than at lower and higher temperatures tested (Table 1). If olive fruit persists in the tree during the cooler months of the year, olive fruit fly can survive and develop in the fruit. When temperatures were increased to 26°C the immatures completed development in the fruit in about 2 wk (Fig. 5), but at 25°C the adults only lived for ~3 wks when provided with food and water in the laboratory (Table 1). When the adults were placed under similar temperatures during the day in the greenhouse environment, longevity was increased to several months (Fig. 1). In related studies, the optimum temperature for development of immature stages in diet was 27°C (Genc and Nation 2008). Maximum diurnal temperatures of ~26°C in this study represent the most favorable weather and climate conditions for rapid development of the immature stages in fruit and maximum longevity of the adults. In addition, the finding that females can produce large numbers of eggs for about 3 wk (Fig. 4) shows the capacity for olive fruit fly to develop very high populations under such favorable conditions as occur during the fall months when mature fruit is abundant.

Survival of olive fruit fly pupa to the adult stage was lowest at 26°C and 41% humidity (Table 3), even though the time from puptation to adult emergence was shorter than the lower temperatures tested. The third instar usually pupates in the soil, so ground temperature will affect developmental time and emergence of adults. Orsini et al. (2007) suggested that reduced eclosion rates were caused by desiccation at temperatures of 31°C and low soil moisture during August. Under favorable weather conditions, the pupal stage is probably the most vulnerable of olive fruit fly life stages because it is not protected in the fruit and does not have the mobility of the adults. However, control tactics directed to the pupal stage would be logistically difficult because of the coverage needed on the orchard floor.

The basic information we developed for olive fruit fly in California in relation to physical and biological factors that affect the survival and development of each life stage will be relevant to assessments of long-term adaptation and population changes under California conditions. Similarly, an extensive amount of knowledge has been accumulated concerning the biology and ecology of Queensland fruit fly, B. tryoni (Froggatt), in Australia (Clarke et al. 2011). The availability of information concerning the effects of physical factors, such as temperature on the life history, development, and reproduction of all stages of this pest, has enabled the development of predictive models based on weather (Yonow et al. 2004). Application of the known genetic changes that have occurred in Queensland fruit fly populations over time (Gilchrist and Meats 2010) implies such changes can also occur in the new California invasion, further separating the U.S. olive fruit fly from the Mediterranean subpopulations.

Acknowledgment

The author is grateful to Gina T. Miller for help in conducting the research and reviewing the manuscript, and to Gail E. Sergent for assistance with experiments. This research
was funded in part by the California Olive Committee, Fresno, CA.

References Cited


GraphPad Software. 2007. GraphPad Prism, version 5.00. GraphPad Software, San Diego, CA.


Received 2 August 2011; accepted 17 October 2011.