

# CALIFORNIA OLIVE COMMITTEE

Interim Reports Submitted for Studies

Funded for the Year 2009

Assembled and formatted by

Marshall W. Johnson  
UC Liaison Officer to the COC

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

Project Year: 2009

Anticipated Duration of Project: 2 Years

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**Project Title:** Release and evaluation of imported natural enemies of the olive fruit fly

### PROJECT SUMMARY

Research objectives are to evaluate imported parasitoids for biological control of olive fly (OLF), *Bactrocera oleae* (Rossi). Two African parasitoids, *Psytalia lounsburyi* (Silvestri) and *Psytalia humilis* (Silvestri) have been approved through previous quarantine evaluations for field release in California. To improve the predictability of this program as to which parasitoid species or its populations may have the best chance of establishment and success in California, we continually (1) release and monitor both parasitoids in the field, and (2) comparatively evaluate key biological traits (thermal performance, overwintering biology, etc.) as well as other factors that may affect the establishment of the parasitoids in California.

#### 1. Field release and monitoring of OLF parasitoids

Field-cage tests have been conducted since 2006 at the UC Kearney Agricultural Center (KAC), Parlier; the UC Citrus Research Station, Lindcove; San Luis Obispo (SLO); and Napa /Sonoma counties, California, to evaluate the effectiveness of *P. lounsburyi* and *P. humilis* on different sized fruit and throughout seasons. In 2009, three field-cages tests were conducted at KAC and Napa. Both parasitoids successfully attacked larval OLF throughout the seasons in the central valley, as well as central coast and Northern California. Mean parasitism levels ranged from 6.2 to 27.4% by *P. lounsburyi*, and 31.3 to 60.3% by *P. humilis*. Within each parasitoid species, parasitism of larval OLF was always higher on the small than large fruit. Overall, parasitism levels of larval OLF by *P. humilis* were higher than *P. lounsburyi*, regardless the fruit size, test dates and locations. This study demonstrates the potential of both parasitoids to suppress OLF populations in California, particularly on small fruit cultivars.

Because high OLF populations occur especially in climatically-mild coastal areas of California, field releases of both parasitoids were thus conducted in SLO and Sonoma, California. In SLO, *P. humilis* was released last year and recovered from the same year, but was not found during early of the sampling of August 2009. This year, about 500, 1000, and 1000 *P. humilis* were released in September, October, and November at three non-commercial olive tree sites, respectively. About 300 *P. lounsburyi* were also released in another site in September. We purposely conducted these releases in non-commercial properties and invited growers or residents to participate the activities. Some growers realized that without a permanent establishment of these introduced parasitoids to reduce the abundant unmanaged fly populations in the neighborhood; their efforts to economically suppress the fly populations in commercial orchards using GF-120 could be limited. In Sonoma, 500 *P. humilis* parasitoids were released in September and October respectively. Post-release samplings from this year's releases in SLO and Sonoma are still under evaluation.

#### 2. Thermal performance

Laboratory experiments were conducted at a series of 9 different constant temperatures (low: 10, 12, 14°C; middle: 18, 22, 26 °C; high: 30, 32, 34 °C) to compare the effects of temperature on the development, survival and reproduction of *P. lounsburyi* and *P. humilis*, as well OLF. OLF failed to develop at 32 and 34 °C, but slowly developed at 12 °C. Both parasitoids could not successfully develop at 10, 12, and 34 °C, but *P. humilis* developed at 32 °C. The suitable temperature range for the development and survival of all stages is 12-30°C for OLF, 14-30°C for *P. lounsburyi*, and 14-32 °C for *P. humilis*, and for the reproduction is 14-32 °C for OLF,

14-30 °C for *P. lounsburyi* and 14-34 °C for *P. humilis*. Thus, OLF is relatively more cold tolerant than both parasitoids while *P. humilis* is relatively more heat tolerant than *P. lounsburyi*. However, mature OLF larvae could develop at 12 or 32 °C. At the constant low (10 °C) or high temperatures (> 32 °C), the flies died mostly during the young larval stages. The survival of endoparasitic and koinobiont parasitoid rely on the survival of its host, and both parasitoids have an advantage by attacking mature larvae that could survived at 32 °C. The studies suggest that there are divergences in thermal performance between the parasitoids and OLF, and there are differences between different these two different parasitoid species. Experiments at variable temperature regimes (10-15 °C, 15-32 °C) are still in progress. Finally, these data will be used to determine the key biological parameters (e.g. low and high thresholds) and temperature-driven relationships for development, survival and reproduction for the development of the host-parasitoid population dynamic models in order to predict the parasitoids' distribution and impact on OLF in California.

### **3. Overwintering biology**

There is a large gap in time when susceptible olives are unavailable to OLF in California (e.g. from early spring to summer). Knowledge on how parasitoids may survive this gap would help understand the colonization biology of these parasitoids in California. Laboratory and field studies were conducted to (1) investigate the effect of low temperature exposure on the parasitoid's fitness (development, survival and reproduction) and how those parasitoids might be able to survive the winter in California; and (2) the effect of cold storage on the parasitoids. Methods of cold storage of parasitoids to induce, maintain, and terminate dormancy in the laboratory have important implications for the use of the parasitoids in biological control programs, which may help develop an efficient method for maintaining fruit fly parasitoid colonies during the non host-fruit seasons, so that a parasitoid colony could be continued for rearing and experiment.

#### **3.1. Low temperature effects on the survival and reproduction of *P. humilis***

Low temperatures reduced the longevity of adult *P. humilis*. The parasitoids survive shorter at either 6 °C or 8 °C than at 10 °C, and males lived shorter than females in all low treatments. On average, female *P. humilis* lived 22, 41 and 54 d at 6, 8, and 10 °C, respectively. During the 2008 winter test, the parasitoids outdoor survived as long as those inside the room; the longest female outdoor died on 16 March, 2009 (i.e., survived 109 days). Although all parasitoids were provided with honey and food, they were not active under < 10 °C, and unable to feed on honeydew. However, the mean winter temperature outdoor was also < 10 °C, but the parasitoids were observed to readily fed on the honeydew during the warming period of middle day. This might have helped them to sustain their life longer than at constant low temperature conditions.

One-week exposure to low temperature (6, 8, 10 °C) appeared not affect the parasitoid's post-exposure fecundity. The fecundities between 8 and 10 °C treatments were similar for all exposure durations, and were higher than those at 6 °C when the exposure periods were > 2 weeks. The parasitoid's fecundity decreased with increased exposure time to each low temperature, and became very weak after a six-week exposure; some of them could not attack the hosts.

#### **3.2. Cold storage effect on the immature development of *P. humilis***

No parasitoid developed after being stored at 6 °C or 8 °C for > 2 months or at 10 °C for > 3 months. Regardless of storage temperature, not a single egg developed after being exposed for > 1 month. A higher percentage of parasitoids emerged from 10 °C than either 6 or 8 °C treatment. Mature larvae or pupae appear to be the most suitable stages for the cold storage. About 48% of emerged adults from the pre-adult treatments (mostly from 10 °C treatment) had deformed wings, and those deformed parasitoids only lived  $3.5 \pm 0.55$  d ( $n = 16$ ). On average ( $n = 15$ ), females developed after being stored at 10 °C survived  $54.6 \pm 8.5$  d with food and water, and reproduced  $2.4 \pm 0.49$  offspring within one week, while normal females (directly reared at  $24 \pm 2$  °C) survived  $65.5 \pm 4.9$  d ( $n = 30$ ), and produced  $15.3 \pm 1.06$  offspring at same conditions. However, developmental times from various immature developmental stages to adults were similar to those parasitoids kept all the time at  $24 \pm 2$  °C.

#### **3.3. Cold storage effect on the survival and development of *P. poneraphaga***

Parasitized OLF puparia by *P. poneraphaga* were stored at 6°C for different periods of time, and then held at controlled room conditions ( $22 \pm 2$ °C, 16L: 8D, 40% RH) at UC Berkeley quarantine. In total, 33 exposures,

ranged from 41 to 94 d, each consisted of 35 to 359 puparia were tested. Developmental time increased while emergence rate decreased with increasing duration of cold storage. Males developed faster than females. Overall, the emergence rate by *P. poneraphaga* (average 17.2%) was considerably higher than that of *P. humilis* being stored at the same cold storage temperature at KAC.

### **3.3 Winter survival and development of *P. humilis* in the field**

Lab-parasitized fly puparia containing *P. humilis* eggs were placed outdoor, once every week from October to December 2008. The parasitoid eggs laid in early October developed into adults within 2008 (developmental time ranged 30-50 d), but those laid in late October developed in January or February 2009 (> 60 d). No parasitoids emerged from the exposures later than 27 October 2008, but unparasitized fly puparia developed into adults in early March 2009. Many parasitized fly puparia were dead, apparently due to the dehydration. Thus, experiments with improved methods by burying parasitized puparia in the soil to reduce dehydration were conducted in the winter 2009 both at KAC and Berkeley I&Q, and those tests are still in progress.

## **4. Other studies being continually conducted in 2009**

### **4.1. Factors affecting sex ratio of *P. lounsburyi***

*P. lounsburyi* is often male-biased in laboratory culture. Experiments were conducted to determine if the number of mating, host stages, host density, temperature or parasitoid density would affect the parasitoid's offspring sex ratio. Preliminary results showed that parasitoid density had a significant effect on the sex ratio. Offspring was female-biased when females were individually reared but male-biased when females were kept in groups during their exposure to hosts, regardless of other conditions tested.

### **4.2. Potential interactions between *P. humilis* and *P. lounsburyi***

To determine competitive outcome between *P. humilis* and *P. lounsburyi*, infested fruit were exposed to each parasitoid alone or one after another in succession. Parasitism by *P. humilis* was five times higher than that of *P. lounsburyi* in single species exposure. The successive exposures of previously exploited hosts by *P. humilis* to *P. lounsburyi* did not significantly reduce the parasitism by *P. humilis*, while the successive exposures of previously exploited hosts by *P. lounsburyi* to *P. humilis* significantly reduced the parasitism by *P. lounsburyi*. However, dissections suggest that both parasitoids have the chance to win intrinsic competition, depending on which parasitoid hatch first in multiple parasitized hosts as the competitive outcome is determined through physical combat among young larvae. Thus, it was possible that above results may result from low parasitization efficiency by *P. lounsburyi*, and detailed studies on the underlying competitive mechanism are still in progress.

### **4.3. Effects of rearing hosts (sterile versus non-sterile hosts) on the fitness of *P. humilis* (in collaboration with Dr. Victoria Yoloyama, USDA-ARS-SJVASC).**

Experiments were conducted to determine the most suitable irradiation doses for the rearing of *P. humilis* on irradiated medfly larvae. The parasitoids were reared on medfly larvae irradiated using five different doses (0, 40, 50, 60, 70 Gy), and the fitness of both immature parasitoids (developmental time, survival) and adult parasitoids (sex ratio, body size, longevity, fecundity, and ability to withstand adverse weather conditions, flight ability) were measured. This study is still in progress.

### **4.4. Influences of honeydew and fruit fly bait on the survival of OLF and parasitoids**

The influences of black scale honeydew and GF-120 bait, as potential food resources, on the survival of adult OLF and two parasitoids, *P. humilis* and *Scutellista caerulea* (Fonscolombe) (a black scale parasitoid) were investigated. In choice tests, *B. oleae* did not show a significant preference between blank GF-120 bait and honeydew, while both parasitoids readily chosen and fed on honeydew. Furthermore olfactory meter tests confirmed that the parasitoids were not attracted by the GF-120 bait. Feeding on honeydew significantly increased the longevities of all three insect species, particularly the parasitoids. Presence of honeydew significantly reduced the fly's mortality when they were exposed to GF-120. The results suggest that presence of black scale honeydew in olive orchards benefits both the pest fruit fly and parasitoids, through providing carbohydrate sources for those insects, but can potentially reduce the efficacy of GF-120 on the fruit fly due to food competition.

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

Project Year: 2010

Anticipated Duration of Project: 1 year

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## **Project Title: Developing Mechanical Harvesting for California Olives: 2007-2010**

### **PROJECT SUMMARY: 2009 Crop Year**

#### **Introduction:**

Our 2009 research produced results in four areas: harvester evaluation, mechanical pruning, orchard establishment and abscission chemical screening. All results were severely compromised by poor fruit set.

#### **Harvester Evaluations:**

All harvesters were tested in a 9 year old hedgerow 'Manzanillo' orchard (202 trees/ac) with a midday leaf water potential (LWP) of -16 bars (well irrigated status) and average fruit removal force (FRF) of 1.68 lbs.

Three trunk shakers, ENE I, ENE II and a Spanish Noli, and one Agright over the row canopy contact harvester (double picking heads) were evaluated. The Two ENE trunk shakers produced final fruit removal efficiencies of 42% and 64%, with 95% and 96% cannable fruit valued at \$1,153 and \$1,146 per ton. The Noli trunk shaker averaged 73% efficiency with 94% cannable fruit valued at \$1,147 per ton. These cannable percentages and values were insignificantly different from hand-harvested fruit values of 97% cannable valued at \$1,178 per ton. Within the course of the experiment trunk damage was largely eliminated by decreasing trunk-clamping strengths to < 1000 PSI and incorporating additional padding between the pad and tree.

The Agright harvester was 68% efficient and produced fruit with a 94% canning percentage valued at \$1,072 per ton; again insignificantly different from hand harvested fruit. However, the harvester produced significant tree damage, as trees could be no more than 10'6" and 6'7" wide and present a straight trunk below 3'6". Trials with this harvester will not be continued.

#### **Mechanical Pruning Evaluations:**

Mechanically topped and hedged trees in a 12 x 26, 14 year old 'Manzanillo' orchard produced an average of 246 lbs/acre with a caning percentage of 96% valued at \$1,174/ton versus 589 lbs/acre with a caning percentage of 94% valued at \$1,157/ton for conventionally pruned trees. The 2009 yield difference was not statistically significant. This is the second year of an 8-year trial.

### **High Density Orchard Establishment Evaluation:**

Three training treatments, a free, a woven and a tied espalier, produced 4.27, 2.22 and 4.39 tons/ac versus 3.46 tons/ac for conventionally trained control trees. These yields, cannable percentages and fruit values were insignificantly different among the four training treatments. This training trial will be discontinued, as the trees will now be pruned to maximize harvester performance.

### **Abscission Chemical Treatments:**

Eight potential compounds were screened; only one demonstrated a significant decrease in fruit removal force. This portion of the project will be discontinued until a more focused approach can be formulated

### **Conclusions**

The overall objective of this project is to develop economically feasible mechanical harvesting that can produce commercially acceptable California black ripe processed table olives.

The primary limiting factor, consumer acceptability has been eliminated by Dr. Guinard's 2009 evaluation of 2008 harvested olives demonstrating neither trained panels nor consumers could distinguish mechanically harvested (canopy contact head) from hand harvested olives. The 2009 receiving station cannable percentages and adjusted prices per ton, insignificantly different from that of 2009 hand harvested control fruit, strongly indicate results similar to the 2008 fruit sensory and consumer panel analysis will be obtained when the 2009 trunk shaker harvested fruit is processed and submitted to sensory and consumer panels in 2010. If this prediction is correct by August 2010 conclusive evidence that both picking methods, trunk shaking and canopy contact head harvesting, produce commercially competitive California black ripe processed olives.

Tree damage was also a limiting factor for both trunk shaking, and to a lesser extent, canopy contact harvesters. For trunk shakers, this 'barking' damage was largely eliminated in 2009 by decreasing clamp strength, adding padding and shake pattern adjustments. For canopy contact heads the branch breakage is not a large problem and can be eliminated by selective pruning before, during and after harvest.

Dr. Karen Klonsky has calculated a 5 ton/ac crop, costing 450/ac to harvest with 80% efficiency, valued at 750/ton, will net a better return than hand harvesting at current hand harvest costs. Therefore, we need to achieve at least 80% final harvester efficiency.

The major remaining problem is improving harvester efficiency, and to a lesser extent, developing the associated machine. Both trunk shaking and canopy contact harvesters have removal efficiencies around 64-68%. Our 2010 research will focus on improving both the harvesting technology and the tree architecture in an effort to increase removal efficiency.

Trials in 2010 will be done cooperatively with ENE Inc., trunk shaking, and Oxbo Corporation, canopy contact harvesters, in more carefully prepared hedgerow orchards in Tulare, Colusa and Glenn counties. The primary objectives will again be evaluating removal and final removal efficiencies, cannable percentages, adjusted prices per ton, and processed fruit quality as evaluated by consumer and sensory panels. Hopefully, we will also be able to develop better assessments of harvester MPH, seconds per tree, tons per hour capacity and harvest price per acre and per ton that will allow growers to assess the value of mechanical harvesting in their orchards.

If successful, 2010-2010 will be the final year of this 4 year project.

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

**Project Year:** 2008 – 2009

**Anticipated Duration of Project:** 3 yrs

**Project Leader:** Dr. Walter Douglas Gubler

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**Project Title:** Implement Diagnosis, Epidemiology, and Control Methods for Fungal Pathogens Causing Stem Canker and Branch Dieback of Olive Trees in California

### PROJECT SUMMARY

Canker and branch dieback of olive trees (*Olea europaea* L.) is a major concern throughout the main olive-producing areas in the Mediterranean regions of Spain, Portugal, Italy, and Greece. The disease is responsible for significant economical losses in mature olive orchards due to both yield reduction and increase of production costs. Increase in production costs are a result of additional cultural and chemical preventive measures as well as diseased wood removal from the infected trees. The California olive industry represents 99% of the total olive production in the United States. In addition, the development of new high density planting olive tree varieties, which can be efficiently machine-harvested, has led to a significant number of new olive plantings and consequently to a notable increase in olive production in California. On the other hand, little is known in regards of “olive branch dieback” and the impact that this disease causes in California olive production. Therefore, the goal of this project was to determine the incidence and importance of “olive branch dieback” in California’s processing orchards and identify the major fungal pathogens associated with canker and dieback disease in both mature orchards and young plantings of olive trees in the state.

In order to determine the incidence and importance of “olive branch dieback”, field surveys were conducted throughout the main olive-production regions in California from October 2008 to September 2009. To date, field surveys have been conducted in 15 different counties including Tehama, Glenn, Butte, Napa, Sonoma, Yolo, Solano, Sacramento, Merced, Madera, Fresno, Tulare, Riverside, Santa Barbara, and Ventura. Symptoms of affected trees were characterized by abundant twig and branch death. Death of the twigs was frequently associated with cankers that extend through the stems and continue through the main branch. In most cases, cankers were found to develop from pruning wounds on the main branches causing eventual death of the productive tree parts. Cankers were also observed in branches and trunks with mechanical injuries as well as natural openings. In addition, cankers were also associated with olive knot galls caused by the bacterium *Pseudomonas syringae* pv. *savastanoi*. Cankers often were observed to colonize the main trunk causing the slow decline and collapse of the whole tree.

In all, over 700 samples of cankered twigs, branches, and trunks were collected from 60 different olive orchards representing the most important varieties planted in California. “Olive branch dieback” was observed in all orchards surveyed in California. However, incidence of the disease varied with location and age of the trees. Results from the isolations of cankered twigs and branches showed fungal species in the family Botryosphaeriaceae, Valsaceae, Diatrypaceae, Coridaceae, and Schizophyllaceae to be associated with olive cankers in California. Botryosphaeriaceae spp., which were isolated from over 30% of the total of samples collected, were the most prevalent fungi isolated from perennial olive cankers and were found in all counties surveyed. In order of importance, Botryosphaeriaceae spp. were followed by Valsaceae spp. (6%), mostly isolated from olive orchards in the San Joaquin Valley and southern California, Diatrypaceae spp. (5.2%), and Basidiomycetes spp. (4%), which were mainly found in olive orchards in the north Coast and the Sacramento Valley. In addition, several mitosporic fungi such as *Aspergillus* spp., *Alternaria* spp., and *Epicoccum* spp. were isolated from cankers but these are considered to be saprophytes. Morphological studies along with DNA analyses of three genes (internal transcribe spacer region ITS1-5.8S-ITS2, a partial sequence of the beta tubulin gene BT2, and a part of the translation elongation factor 1- $\alpha$  gene EF1- $\alpha$ ), allowed us to identify the most predominant fungi isolated from olive cankers to species level. To date, we have identified 8 different species within the Botryosphaeriaceae family including, *Botryosphaeria dothidea*, *Lasiodiplodia theobromae*, *Diplodia mutila*, *Diplodia seriata*, *Dothiorella iberica*, *Neofusicoccum australe*, *Neofusicoccum mediterraneum*, and *Neofusicoccum vitifusiforme*; 3 species within the Diatrypaceae family including, *Eutypa lata*, *Diatrype* sp., and *Diatrypella* sp.; 3 species within the Valsaceae family including *Cytospora* sp., *Phomopsis* sp., and *Diaporthe ambigua*; and 2 Basidiomycetes spp. including, *Trametes versicolor* and *Schizophyllum commune*.

Previous studies conducted in our laboratory have shown species in the Botryosphaeriaceae and Diatrypaceae families to constitute the primary cause of cankers and consequent dieback on grapevine in California. Therefore, their common occurrence in olive trees may suggest that these fungi may also be associated with olive branch dieback as well. However, whether or not these fungi play an important role as primary pathogens causing olive branch dieback is still not clear at this time. Consequently, Koch’s postulates are currently underway to determine which fungal species isolated from cankers of olive trees are the main pathogens causing dieback of olive in California. Pathogenicity tests are currently ongoing in Manzanillo and Sevillano trees at the Nickels Soil Laboratory in Arbutle, California. Control using properly timed fungicide applications immediately after pruning and or harvesting are being established.

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

Project Year: 2009-2010

Anticipated Duration of Project: 1 year

**Project Leader:** Art Hutcheson

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**Project Title:** Development and Performance Evaluation of the Hutcheson Mechanical Olive Harvester

**Cooperators:** Louise Ferguson and Uriel Rosa

**Reporting period:** February 10, 2009 – November 3, 2009

### PROJECT SUMMARY

#### Introduction:

The goals I started with were to build a machine that was:

- Economical for the average grower
- Smaller in design than the current Korvan type machine yet able to do full size trees
- Able to mimic a hand picker in its harvesting action rather than knocking the fruit from the tree
- At minimum replace the daily output of a 50 man hand crew
- Harvest olives with quality equal to or better than the hand crew

The current machine was built to wrap around one tree at a time. An operator then moves the picking head over one side of the tree cleaning it completely before moving on to the next tree. Two men are required to operate the harvester.

#### Objectives:

During a brief test period in October of 2008, I observed several deficiencies in the design of the picking arm and hydraulic system which I plan to correct this year.

The following objectives are needed to resolve the deficiencies found in during the test period:

1. Improve functions of current picking arm, both hydraulically and mechanically
2. Improve padding and metal covering to avoid damage to falling fruit
3. Put to use under harvest conditions in a commercial orchard with the help of Louise Ferguson

#### Procedures:

In June the six tree rows assigned to mechanical harvest will be skirted to three feet.

In October both sets of experimental trees will be harvested:

- The 6 hand harvested rows will be harvested by hand, and the total row weight taken.
- The bins from each lot will be combined into a single lot.
- The bins will be graded as a single lot at the receiving station.

The 6 machine harvested rows will be harvested and the total row weight taken.  
The time to harvest the six rows will be recorded to determine harvest time per tree.  
The bins will be combined into a single lot.  
The bins will be graded as a single lot at the receiving station.

If significant fruit remains on the ground after the machine harvest, it will be collected and weighed.

The machine harvested trees will be hand harvested and weighed.

The following calculation will determine the efficiency of the mechanical harvester:

Weight Machine Harvested

(Weight of machine harvest + hand gleaned fruit weight) = harvester efficiency

The receiving station grade of the two, six row sets will be statistically analyzed to determine the effect of harvesting on fruit quality and grade.

The analyzed data should give:

1. The harvest efficiency of the mechanical harvester.
2. The harvest time per tree.
3. The effect of machine harvest on fruit quality.

This experiment can be done with different tree spacing or tree heights.

## **Results and Discussion:**

As described in the objectives above mechanical and hydraulic improvements were completed to picking arm, padding was added, and our test plot at Cilker Orchards was prepared to Louise Ferguson's specifications.

On October 4, 11, and 18<sup>th</sup> 2009, the harvester entered the orchard to do some trial runs and it was apparent that the picking head itself had some problems. As it entered the perimeter of the trees the fruiting branches would wrap around drum so tightly as to break and in one case a 3 inch scaffold was pulled from the tree. After some modifications to the head, I tried again with much the same result.

## **Conclusions:**

On October 19<sup>th</sup>, Louise Ferguson, Uriel Rosa, and Steve Sibbett arrived to observe. Uriel suggested running the pick head in reverse and at a different angle. This helped somewhat but we still had a problem with branch wrapping. We discussed welding some end plates on the drum and stopping the rotation using the head in a rake motion. The other suggestion was to change from a rotating drum with 4 rows of fingers to a single rake or comb type mechanism. During all of these trials not enough fruit was harvested to make any efficiency or quality evaluations. As such, I would like to extend this project one more year to correct these new problems.

Since we have some Navadillo olives that we harvest for oil in December and January, I will be trying to harvest with Uriel's suggestions at that time.

## **Funding Sources:**

Art and Justine Hutcheson and the California Olive Committee

**CALIFORNIA OLIVE COMMITTEE**  
**Interim Progress Report 2009**

**Project year**   2010                        **Anticipated Duration of Project**   One Year  

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**Project Title:** Appraising the Threat of Olive Psyllid to California Table Olives

**PROJECT SUMMARY**

The olive psyllid, *Euphyllura olivina* (Costa), was discovered in July 2007 in the California counties of San Diego and Orange. Nymphs and adults feed by rupturing cells and ingesting sap, reducing nutrients that are essential for tree development and fruit production. Plant parts attacked include the buds, tender shoots, floral axes, inflorescences, and young fruits. Nymphs secrete a sticky wax (similar to mealybugs), which accumulates on the foliage and stimulates premature flower drop. When colonies are on inflorescences, the level of sap loss is directly proportional to psyllid numbers. Yield reductions can be as much as 40 to 60%. Funding was provided by the COC in 2009 to address the following objectives.

**Objective 1.** Determine and map the distribution of *Euphyllura olivina* over the next 12 months and quantify infestation levels within California.

**Results.** Prior to COC funding in 2009, initial surveys were conducted in July and October 2008 at more than 20 sites in the counties of San Diego, Orange, Los Angeles, and Riverside. Five surveys during March to October 2009 in the same counties were funded by the COC and included more than 40 sites that were revisited to document the population trends of the psyllid at the various locations. Olive trees in residential (around apartment complexes, homes), business (malls, shopping centers, parking lots, etc.), and agricultural areas (near vineyards, roadside landscapes) were randomly selected and inspected for olive psyllid colonies and the wax they produce (the psyllids within the waxy mass was termed a “cluster”). At several sites the psyllid’s presence (as indicated by waxy formations resembling cotton balls) was easily detected by the untrained eye. For each survey in 2009, between 341 and 624 trees were checked among the sites (**Fig. 1**). At each site, each tree was given an infestation rating of 0 to 4 (0 = non-infested; 1 = 1 to 3 psyllid clusters; 2 = 4 to 10 psyllid clusters; 3 = light infestation with 1/10<sup>th</sup> of tree infested; 4 = heavy infestation with more than 1/10<sup>th</sup> of tree infested). The current olive psyllid distribution appears to include areas from Huntington Beach, Newport Beach, and Irvine (Orange County) in the north to the city of San Diego in the south and east to Escondido, near Fallbrook, and Temecula (**Fig. 1**). To date, olives checked in Palos Verdes, Anaheim, Burbank, Santa Clarita, Sylmar, Riverside, Sun City, Perris, and Hemet were free of the psyllid.

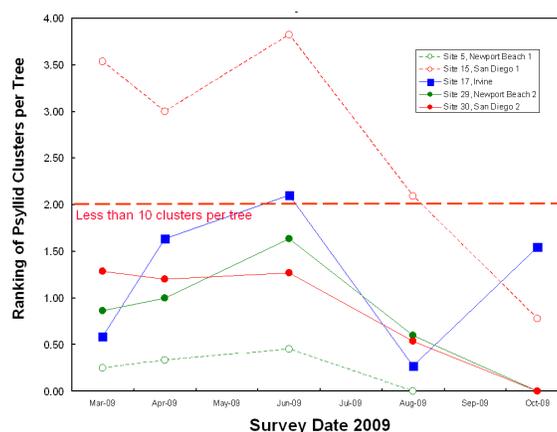
The percentages of trees with olive psyllid within the infested areas varied among sites and the time of the surveys (i.e, March, April, June, August, & October). For example, in June 2009, the psyllid was found on 62.8% of the trees within the infested areas. At the majority of sites, usually more than one tree would be infested even when infestations were very low (e.g., 1 to 3 clusters per tree). This suggests that once the psyllid becomes established in a location, the adults disperse to many trees in the vicinity. However, only in a few localities (Carlsbad and San

Diego) were the populations considered potentially damaging (infestation ratings were in the 3's and 4's). From October 2008 to June 2009, the percentage of infested trees increased at the checked sites and then percentages dropped in August and October 2009 following the summer period (Fig. 2).

When originally surveyed in 2008, *Euphyllura olivina* infestations were confined to an area within 15 miles of the coast in eastern San Diego and Orange Counties. In 2009, it was found for the first time east of Fallbrook near I-15 (San Diego County) and at three locations around Temecula (Riverside County). These areas are about 30 miles from the coast. If *Euphyllura olivina* can survive the summer temperatures in the Temecula area, it may be able to survive in the Central Valley. A continuation of the survey is needed to evaluate the potential for the olive psyllid to survive and expand in the Temecula area and beyond to the Central Valley.



**Fig. 1.** Sample sites in southern California where olive psyllid was found infesting olive trees in urban and agricultural areas in the counties of San Diego, Orange, and Riverside.



**Fig. 2.** Changes in mean ranking of psyllid cluster infestations at five survey sites in southern California from March to October 2009.

**Objective 2.** Identify and quantify the impact of existing natural enemies (e.g., lacewings, ladybugs, parasitoids, etc.) that are attacking *Euphyllura olivina* stages in infested areas.

**Results.** Over the survey period very few natural enemies were seen in association with olive psyllid clusters on infested olive plants. Green lacewing larvae, mealybug destroyer (*Cryptolaemus montrouzieri*), and some ladybugs were infrequently found associated with the olive psyllid. Three times large numbers of psyllid infested branches were collected from several heavily infested sites and held in the laboratory (UC Irvine and UC Berkeley quarantine) to see if parasitoid natural enemies would emerge from the collected material. No parasitoids were found in association with the psyllid.

A parasitoid, *Psyllaephagus* spp. (Hymenoptera: Encyrtidae), was collected in Spain by K. M. Daane, UC Berkeley, and C. Pickett, CDFA, and shipped to the quarantine facility at UC Berkeley. This species was maintained in colony in the quarantine for several months, but was lost due to problems in establishing a continuing olive psyllid colony in the quarantine. Adult female psyllids did not lay eggs on the olive plant material provided to them. Because of this, no nymphal stages were available for the parasitoid to parasitize and the adult *Psyllaephagus* spp. finally died out. Tentative plans are being made by Daane and Pickett to collect the parasitoid again in Spring 2010.

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

Project Year: 2009-2010

Anticipated Duration of Project: 12 months

**Project Leader:** Rodrigo Krugner

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**Project Title:** Evaluation of pathogenicity and insect transmission of *Xylella fastidiosa* strains to olive plants

### PROJECT SUMMARY

*Xylella fastidiosa* (*Xf*) is a xylem-limited bacterium that causes a number of diseases in economically important crops in California and worldwide. In California, *Xf* strains are transmitted by species of leafhoppers, including the glassy-winged sharpshooter (GWSS), *Homalodisca vitripennis*, which has now established in portions of the San Joaquin Valley and most of southern California. Because it is a highly mobile and polyphagous leafhopper, with over 100 known hosts, it changed the epidemiology of diseases caused by *Xf*. About 145 plant species have been identified in California as potential hosts of *Xf* strains, including olive plants.

In recent years, scorching symptoms in olive trees potentially caused by *Xf* infection have emerged as a potential disease threat to olive production areas in California. However, the role of *Xf* as the causal agent of “olive leaf scorch disease (OLSD)” has not been fully investigated. Knowledge of the disease symptoms and genetic diversity of OLSD strains associated with OLSD in California will help in devising effective strategies for managing OLSD.

The objectives of this project are: 1) to increase our understanding of the pathogenicity of *Xf* strains and accurately describe the symptoms in olive plants; 2) to identify potential vectors of *Xf* in olive orchards and to evaluate the transmission efficiency of *Xf* in olive plants by GWSS, and 3) to characterize temporal and spatial patterns of disease incidence within selected orchards and urban areas, and to determine the genetic structure of *Xf* strains associated with the scorching symptoms. An ultimate goal of this research will be to provide fundamental, research-based information needed to manage this potential disease threat in Central Valley orchards. Results from these studies are anticipated to benefit olive growers and crop consultants and will be essential for properly timing any vector control measures and/or implementation of cultural practices.

### PROGRESS REPORT

To date, a total of 90 samples of olive plants showing limb dieback (Fig. 1A) and leaf scorching (Fig. 1B) symptoms were photographed and collected from trees in urban areas in central, coastal, and southern California to determine if *Xf* is constantly associated with the disease. Samples were screened in laboratory by PCR and culturing. A total of 23 samples were PCR positive for *Xf*, from which three *Xf* strains were isolated (Table 1). None of the samples collected in northern California and San Joaquin Valley were PCR

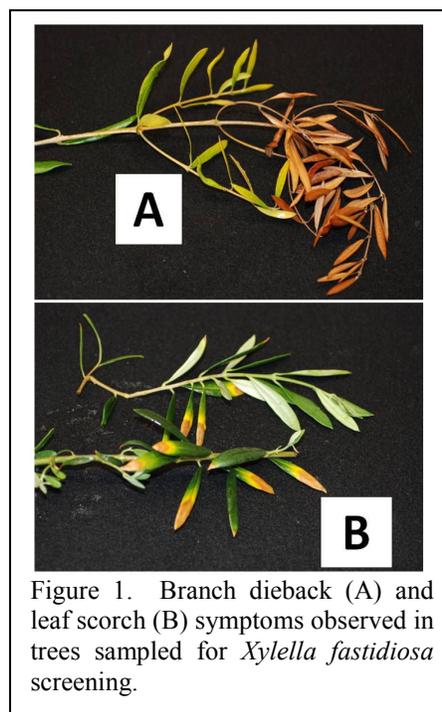


Figure 1. Branch dieback (A) and leaf scorch (B) symptoms observed in trees sampled for *Xylella fastidiosa* screening.

positive for *Xf*, except three samples collected in Ducor. However, those were weak positives that are being retested.

Bacterial cells from the Rolling Hills isolate were needle-inoculated in healthy olive plants (cultivars: Mission, Manzanillo, Sevillano, Mission, Arbequina, Arbozano, Koroneiki, and Barouni) in March 2009 and monitored every 6 weeks for symptom development. Although *Xf* was detected in test plants by PCR, branch dieback and scorched leaf symptoms have not been consistent among inoculated plants. A second group of plants (Manzanillo only) were inoculated on October 2009 with the isolate from Fillmore, which appeared to grow faster in laboratory media than the Rolling Hills isolate.

Isolates of *Xf* obtained from pure cultures and bacterial DNA extracted from symptomatic plants are being characterized by pathogen genotype identification based on single nucleotide polymorphisms analysis from genomic loci and phenotypic analysis of cultured isolates based on in vitro growth characteristics. Preliminary results indicate that *Xf* in all positive samples have the type A genotype, which is known to cause Almond and Oleander Leaf Scorch Diseases, but not Pierce's Disease in grapevines.

**Table 1. Number of olive plant samples collected in California from August 2008 to August 2009 and laboratory screening results for *Xylella fastidiosa*.**

Collection date	Location	Number of samples	PCR result	Isolation result
<i>Northern California – Yolo County</i>				
May 09	Davis	4	0/4	0/4
<i>San Joaquin Valley – Kern and Tulare Counties</i>				
August 08	Bakersfield	5	0/5	0/5
August 08	Porterville	3	0/3	0/3
August 09	Ducor	20	3*/20	0/20
<i>Southern California – Orange County</i>				
May 09	Newport Beach	8	2/8	0/8
May 09	Costa Mesa	7	1/7	0/7
<i>Southern California – Los Angeles County</i>				
October 08	Rolling Hills	5	0/5	1/5
March 09	Rancho Bernardo	3	3/3	0/3
April 09	Rolling Hills	3	3/3	1/3
<i>Southern California – Ventura County</i>				
August 09	Piru	5	2/5	0/5
August 09	Fillmore	7	1/7	1/7
August 09	Ventura	3	3/3	0/3
<i>Southern California – San Diego County</i>				
May 09	Carlsbad	3	3/3	0/3
<i>Southern California – Riverside County</i>				
May 09	Riverside	8	0/8	0/8
August 09	Riverside	6	5/6	0/6
	<b>Total =</b>	<b>90</b>	<b>23</b>	<b>3</b>

\*Weak positive band on PCR assay

# **CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT**

**Project Year:** 2010-2011

**Anticipated Duration of Project:** 3 years

**Project Leader:** Dr. Alyson E. Mitchell

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**Project Title:** Improving California-Style Black Ripe Olive Quality

## **PROJECT SUMMARY**

Traditional California-style processing methods are designed to remove naturally bitter compounds which include many of the phenolic compounds with biological potential. The main phenolics found in olives include oleuropein, hydroxytyrosol, tyrosol, luteolin and rutin. The predominate phenolics in a California-style table olives are tyrosol, while hydroxytyrosol is the main phenolics in other style of table olives. Hydroxytyrosol has been shown to reduce the risk of chronic diseases such as cardiovascular disease and cancer, while tyrosol does not show any appreciable effect. California-style olives have low levels of these phenolics as compared to other table olives as most of the hydroxytyrosol is oxidized during color fixation by ferrous ions (ferrous gluconate).

California-style processing also leads to the formation of acrylamide, a compound involved in cancer development in experimental animals. Traditional California-style black ripe olive processing method consists of a treatment with sodium hydroxide followed by iron-salt treatment and air-oxidation. The method is designed to remove oleuropein, a naturally bitter compound in olives, however, acrylamide is formed during this process.

Acrylamide is usually found in food which undergoes heat processes. Free amino acids, mainly asparagine, and reducing sugars have been suspected to be the main precursors through Maillard reactions. However, the mechanism of the formation of acrylamide in California-style black ripe olives seems to be different since the amount of free amino acids and reducing sugars in raw olives does not correlate to the amount of acrylamide found after sterilization. The precursors as well as the precise mechanism of the formation of acrylamide in California-style black ripe olives are still unclear. The objectives of this project are to understand the mechanism of the formation of acrylamide in California-style black ripe olives and develop novel ways to reduce acrylamide in the final products.

We hypothesize that the addition of a natural source of hydroxytyrosol, derived from olive processing waste materials, to the olives during lye treatment and prior to sterilization and color fixation has the potential to improve the nutritional and functional value of California-style olives without altering flavor. This addition may also have the added benefit of reducing acrylamide levels in California-style olives as the excess

hydroxytyrosol may reduce the formation of acrylamide precursors (derived from lipid oxidation) which form either during lye treatment or ferrous gluconate treatment. We believe it is possible to use olive waste and waste water obtained from processing as an economical source of hydroxytyrosol; reducing processor waste and improving industry sustainability.

## **PROGRESS REPORT**

### **1. LC-MS/MS methods have been developed and optimized for analyzing acrylamide in olives.**

*Extraction methods developed:* Samples consisting of 30 g of olives were extracted with 60 ml of water. A 1 ml sample of 4 ng/g of D<sub>3</sub>-acrylamide, an internal standard, was added to samples. The mixture was homogenized and 15 ml of hexane is added to remove oil and other non-polar compounds. The mixture was centrifuged at 4000 rpm for 30 minutes. Aqueous layer was collected. The solution was cleaned up with Strata-X-C (200 mg, 3 mL) cartridge. Acrylamide was eluted with 1 ml of mobile phase (0.1% formic acid in water/methanol, 90:10, v/v). The eluted solution was collect and analyzed by LC-MS/MS method.

*LC-MS/MS methods developed:* The quantification of acrylamide was performed using LC-(ESI+)/MS/MS using a Prodigy ODS column (100A<sup>0</sup>, 4.6 mm x 250 mm, 5 μm). The mobile phase was the mixture of 0.1% formic acid in water/methanol (90:10, v/v). The LC/MS/MS was optimized using a capillary voltage of 3.0 kV, a cone voltage of 20V, in (+) mode ESI at a source temperature of 140 °C and using a desolvation gas temperature of 400 °C, 550L/hr desolvation gas flow rate. The collision energy for multiple reaction monitoring (MRM) was set at 12eV for both acrylamide and D<sub>3</sub>-acrylamide. Transition ions monitored for quantification were m/z 72 > 55 for acrylamide and m/z 75 >58 for D<sub>3</sub>- acrylamide.

### **2. Information gathering on California-style black ripe olive processing.**

We (Dr. Mitchell, Suthawan Charoenprasert and Dan Flynn) visited Bell Carter on 08/25/09 and met with Julia Workman and Jane Yegge. During this visit we toured the processing facility and learned about how Bell Carter processes California-style black ripe olive. At this time we took samples and also set up sampling protocols for future experiments. We also visited with Musco on 10/02/09 and toured the processing facility, learned about their processing methods, took samples and also set up sampling protocols for future experiments.

### **3. Levels of acrylamide have been surveyed in fresh, Spanish-style, Greek-style, Canned green ripe and California-style black ripe olives**

In order to better understand about the mechanism of the formation of acrylamide in California-style black ripe olives, acrylamide in fresh olives and the four types of table olives as shown in table 1 have been determined. High concentration of acrylamide has

been found in California-style black ripe olives followed by canned green ripe olives, while acrylamide level in fresh, Spanish-style and Greek-style table olive is much lower. California-style black ripe and canned green ripe olives are treated at higher temperature than Spanish- and Greek-style table olives. Canned green ripe and California-style black ripe olive processing method are very similar, but canned green ripe olives are not oxidized during lye treatment, and they are not treated with ferrous gluconate. Therefore, high heat treatment, air oxidation and iron-salt treatment seem to play an important role in the formation of acrylamide in California-style black ripe olives.

**Table 1** Acrylamide levels in fresh, Spanish-style, Greek-style, canned green ripe and California-style black ripe olives

Olives	Acrylamide in olive (ng/g) (wet basis)		
	Sample 1	Sample 2	Sample 3
Fresh olives (Manzanilla)	<LOD	<LOD	<LOD
Spanish-style table olives	<LOD	<LOD	<LOD
Greek-style table olives	<LOD	<LOQ	<LOD
Canned green ripe	90.7±0.7	102±1.2	63.5±0.5
California-style black ripe olives	467±5.0	386±4.8	405±3.0

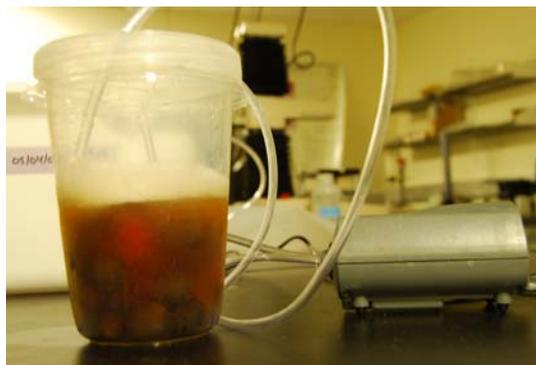
Results are mean values of two measurements

<LOD = Concentration of acrylamide is lower than limit of detection

<LOQ = Concentration of acrylamide is higher than limit of detection but lower than limit of quantification (LOQ = 3.41 ng/g)

#### 4. Developed laboratory scale models for mimicking California-style black ripe olive processing.

It is necessary to have carbon sources in order to form acrylamide. The oxidation of fatty acids in olives during lye and iron-salt treatment may provide acrylamide precursors. Thus addition of antioxidant into the olives during processing could help prevent the formation of acrylamide precursors and help reduce acrylamide level in the final products. To date, we have developed a series of lab-scale models to mimic California-style black ripe olive processing, in order to save costly chemicals such as antioxidants and labeled fatty acids as well as other materials. Air is injected into olive in lye solution and ferrous gluconate solution by air pumps as shown in **Figure 1**. Olives are sterilized in an autoclave at temperature of 250 °F for 30 minutes.



**Figure 1.** Model mimicking California-style black ripe olive processing.

Three types of water including nano-pure, distilled and regular water have been used to prepare sodium hydroxide, ferrous gluconate and brine solution which were used in the models. Olives treated with nano-pure and distilled water were yellow-brown after treated with lye and ferrous gluconate, and the concentration of acrylamide found in these olives was much lower than reported in California-style black ripe olives. Olives treated with regular water contain much higher acrylamide and they became black after treated with lye and ferrous gluconate. Therefore, regular water seems to be more suitable to be used in the model than highly pure water. Optimization of models is on going.

## CALIFORNIA OLIVE COMMITTEE INTERIM RESEARCH REPORT

Project Year: 2009-2010

Anticipated Duration of Project: One year

**Project Leader:** Uriel A. Rosa

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Cooperators: Louise Ferguson, Phil Scott, Erick Nielsen, Sergio Castro-Garcia, Kitren Glozer, Bill Krueger, Jackie Burns, John Ferguson.

**Project Title:** Development of (a) Canopy Shaking and (b) High Frequency Trunk Shaking Machines for Mechanical Harvesting of California Table Olives.

### PROJECT SUMMARY

#### **Project Objectives: 2009**

The goals of this proposal are to improve, implement and evaluate the design of the following available harvest equipment. The modified equipment is intended to harvest high quality table olives in California.

- a) ENE trunk shaker equipped with multi-directional computer controlled Terry Tompkins head.
- b) ENE boom chassis retrofitted with the Noli high frequency orbital shaker imported from Spain.
- c) AgRight over-the-row canopy harvester equipped with padded drum, conveyors and catching frame.

#### **Major Accomplishments:**

a) ENE Shaker:

Extensive hours were spent on improving this design. But we satisfactorily achieved two major goals. First, and very important, we developed a padding system that successfully avoids bark damage. This is especially important, because the trees we harvested were standing on wet soils. Second, new shake patterns were programmed into the computer that controls the two unbalanced rotating masses. Suitable patterns have been tested and implemented by reprogramming the shaking vibration with a laptop computer. Developed shaking patterns are easily selected in the field. Around twelve different olive shaking patterns were created. Some particular patterns showed best results. Additional features include easy adjustment and repeatability of total shake time, smooth start and stop of the head and combinations of patterns during the same shake period.

b) Noli Shaker: We successfully imported this unit from Spain and retrofitted it onto an OMC nut harvest chassis. Fruit removals were similar for these two shakers during these trials, around 65% to 70%.

c) AgRight harvester: The fruit quality observed from this unit ratifies that the canopy shaker principle is adequate to harvest good quality fruit if the harvester is properly padded. The recorded ground speed around 0.25 mph is much higher then compared with previous years, but may be further increased. Note that the entire tree is harvested at this speed, as oppose to previous years.

#### **Important Considerations:**

a) and b)

Adequate pruning needs to be developed for optimum performance of the trunk shakers. Quality of fruit harvested during the 2009 trials has been evaluated. Visual inspection and preliminary results indicate fruit quality is very high. Typical required shaking time may be around 15 to 20 seconds per tree. Pruning and shaker head studies can further improve fruit removal efficiency. It was also

noted that trunk shakers may leave fruits on concentrated spots usually on flexible hangers outside the tree canopy which could be picked with other mechanical methods. Follow up studies could be conducted on this concept.

c)

For over-the-row harvesters, tree size has to match the maximum harvester dimensional capabilities to avoid tree damage. Although the harvester showed average harvest speeds of 0.25mph, it seems that this ground speed could be further increased. Proper pruned trees are required.

### **Brief Description of Redesigned Harvest Equipment**

a) A shaker head was acquired from Gold Country hydraulics and retrofitted onto a ENE trunk shaker unit. A receiver was also used on field operations to complete the catch-shake setup. An innovative computer controlled servo system was developed and tested at the Nickels Ranch.

Among other features, this system can be reprogrammed to produce different excitations and patterns produced by the two distinct vibrating motors. Acceleration and deceleration of the motors are smooth during start and stop of the vibration. Among innumerable tested materials a combination of fill pads and double layer rugs produced excellent results and were adopted as the final padding system. The clamping pressure was adjusted for optimum performance and to avoid bark damage. The adjustment of eccentric masses, possibility of programming and easily selecting numerous vibrating patterns and adjusting vibrating time made this unit quite versatile. Further investigations can be performed with the use of this computer controlled unit.

b) The Noli shaker, imported from Spain, was hydraulic controlled but not computer controlled. A fill pad made of our especially developed Kevlar pads created a large contact area and allowed adequate transmission of vibration to the trees. The kevlar pads were developed in US and have been instrumented and thoroughly tested in Spain by Dr. Sergio Castro. We added two rugs between the trunk and the fill pads, similar to the ENE setup, to protect the delicate wet bark during vibration. The hydraulic pressure of the clamping cylinder was limited to 800 psi for optimum performance and to avoid bark damage. Actual average contact area and bark pressure can still be evaluated during this study. Our initial tests at Nickels Ranch produced bark damage because we did not have adequate rug pads installed neither appropriate clamping pressure set on the hydraulic circuit. After proper adjustment of clamping pressure and introduction of double rugs the bark damage was avoided, even that we only had wet soils during the trials.

c) Olivia, the AgRight modified pomegranate harvester of Dave Loquaci required padding modifications to be table olive ready. The shaking rods, catch frame and other required hard surfaces were padded to reduce fruit damage. The tips of padded rods were equipped with plastic end caps to protect the rod rubber pads against mechanical failure. Our GPS-based Olive yield monitoring system was installed on top of the harvester to record all machine movements and later calculate performance based on harvester ground speed and measured bin weights. The bin scale for this trial was not installed on the harvester. The bin weights were recorded on scales set on the ground. Speed and weight data were recorded and have been analyzed.

## PROGRES REPORT AS OF NOVEMBER 1, 2009

Project year: 2009

Duration of the project: April –November 2009

**Project Leader:** Jim Stewart

Location: Tulare County

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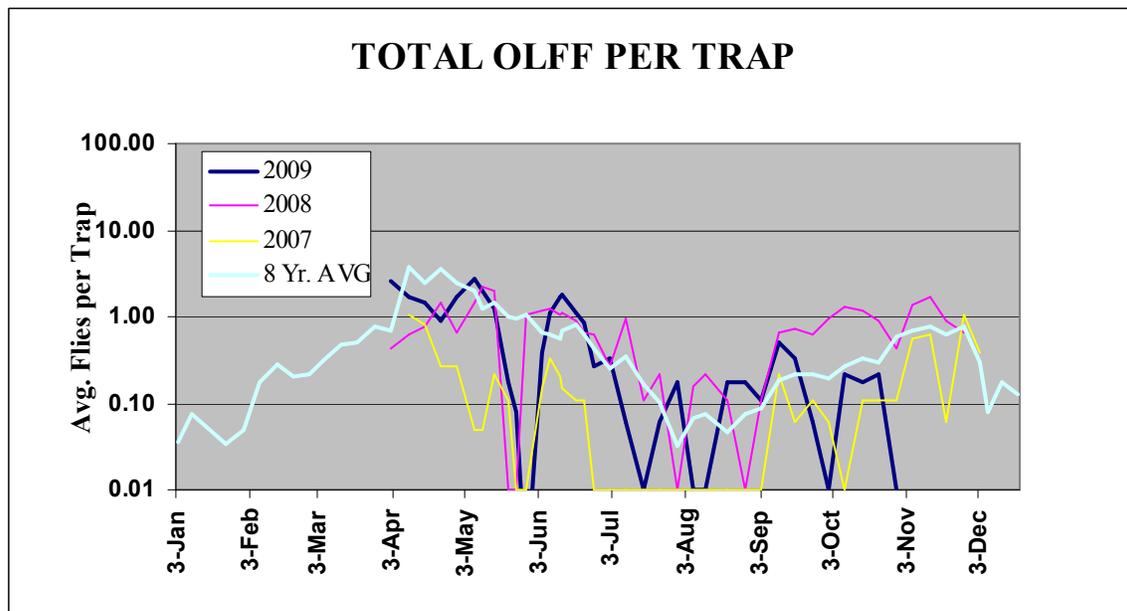
**Project Title:** Southern San Joaquin Valley Olive Fruit Fly Monitoring Project

Cooperating personnel: Bert Quezada, Jamie Nemecek

Keywords: Olive Fruit Fly, Monitoring, Traps,  
Commodity: Olive

### PROGRESS TO DATE (November 1, 2009)

Two traps were set up in each of the 9 groves as detailed in the proposal by April 1 of this year. Traps were read & serviced on a weekly basis. Data was collected and reported by fax and/or e-mail to interested parties. The results indicated that the general population of Olive Fruit Fly (OLFF) was above last year's level but below the 8 year average. Traps of late (October 31) have indicated a drop in the population even though temperatures have been above normal (see graph). Trapping will continue through November.



**CALIFORNIA OLIVE COMMITTEE  
INTERIM GRANT REPORT**

Project Year: 2009-2010

Anticipated Duration of Project: 2 Years

**Project Leader: Victoria Y. Yokoyama, Ph.D.**

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**Project Title: Biological and Cultural Control of Olive Fruit Fly in California---  
Utilization of Parasitoids from USDA-APHIS-PPQ, Guatemala and  
Cultural Control Methods**

**PROJECT SUMMARY**

Research objectives are to a) continue releases of the parasitoid, *Psytalia* cf. *concolor* = *P. humilis*, in different regions in California for biological control of olive fruit fly, b) evaluate other parasitoids that are mass-produced in Guatemala for importation, c) develop methods to rear a domestic supply of large numbers of parasitoids, and d) develop cultural control practices that further reduce pest populations in the inland valley regions.

1. Continue Field Releases of an Imported Parasitoid for Biological Control.

The olive fruit fly parasitoid was mass produced in the USDA, APHIS, PPQ Moscamed laboratory in San Miguel Petapa, Guatemala and imported into the USDA, ARS, SJVASC laboratory in Parlier, CA. Recent genetic studies have shown that the parasitoid is distinct from related species, and the name was changed to *P. humilis*.

Parasitoid releases in late 2008 through January 2009 were evaluated with post-release samples of fruit in regions and locations as follows: Sacramento Valley at Orland and Oroville; North Coast at San Jose; San Joaquin Valley at Lemon Cove; and the Central Coast at Solvang. Subsequent generations of the parasitoid were not recovered from olive fruit fly at these sites primarily due to the scarcity of fruit in the trees and negligible olive fruit fly infestations during the winter months.

Parasitoid releases in 2009 were begun in September with emphasis on olive fruit fly infestations on the east and south side of the central valley including Grapevine, Bakersfield, Porterville, Visalia, Lodi, and Oroville. Olive fruit fly numbers and rates of parasitism in these locations are under evaluation. Prior to a 2009 parasitoid release in San Jose, fruit infested with olive fruit fly were evaluated for new generations of parasitoids that may have resulted from a previous 2008 release.

## 2. Evaluate Mass-Reared Parasitoids in Guatemala for Importation.

Average flight endurance of the parasitoid *P. humilis* ranged from 4-21 and 7-14 minutes for females and males, respectively under different environmental conditions; and the longest flights were about 75 and 39 minutes for the females and males, respectively. The results show that the parasitoids are capable of flight over long distances which are favorable for dispersal among olive fruit fly infestations in different areas.

The effect of rearing parasitoids from irradiated Mediterranean fruit fly host is under investigation. Irradiation provides a secure method of mass producing the parasitoid and prevents accidental contamination of shipments with fertile hosts.

USDA, APHIS, PPQ permits are in process to import two parasitoids, *Fopius ceratitivorus* and *P. corsyrae*, that are mass produced in Guatemala into the UC Berkeley quarantine facility for evaluation for biological control of olive fruit fly. *F. ceratitivorus* was found established in olive fruit fly infestations in the middle east.

## 3. Develop Methods to Mass Produce Parasitoids on Olive Fruit Fly on Lab Diet.

The parasitoid was not stimulated to lay eggs in mature olive fruit fly larvae when the larvae were exposed to the females in diet or in different devices with domed shapes to simulate olive fruit.

The olive fruit fly colony reared on formulated diet has been maintained through 40 generations. The colony will be used for collaborative research with the USDA, ARS, Albany, CA for irradiation studies that are related to development of a sterile insect technique for olive fruit fly.

## 4. Develop Cultural Control Methods for Olive Fruit Fly.

The distance that olive fruit fly adults can travel without flight was at least 20 feet in laboratory tests and the maximum distance is still under investigation. The dispersal capacity of the third instar larvae and the non-flying, newly emerged adult is studied in relation to containment by ground mulches.

A large yellow pan trap was obtained from the USDA, ARS, Hilo, HI for evaluation for control of olive fruit fly. The trap was developed in Hawaii and shown to be effective in reducing numbers of tropical fruit fly species and cherry fruit fly.

## 5. 2009 Publications.

Yokoyama, V. Y., C. E. Cáceres, L. P. S. Kuenen, X. G-Wang, P. A. Rendón, M. W. Johnson, and K. M. Daane. 2010. Field releases, reproduction, and flight endurance of an olive fruit fly parasitoid, *Psytalia humilis* (Hymenoptera: Braconidae), reared on irradiated Mediterranean fruit fly. Biological Control (Manuscript submitted).