CRB-FUNDED RESEARCH PROGRESS REPORT



Photo 1: Yellow sticky cards used for trapping adult psyllids.

COMPARISON OF TAP AND Sweep net monitoring Methods for ACP

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The Asian citrus psyllid (ACP) was first detected in California in 2008. It is widely distributed throughout residential and commercial citrus in Southern California and is now expanding its range into the Central Valley. It is among the most damaging pests of citrus and closely-related plants, primarily due to its activity as a vector of the bacterium associated with huanglongbing (HLB), also known as citrus greening disease.

Proper management of ACP and the HLB-associated bacterium it transmits requires a comprehensive monitoring program to detect psyllids, quantify psyllid densities, define treatment thresholds and determine the efficacy of treatments. Psyllids are prevalent throughout the year in southern California, but peak activity occurs during periods of actively growing flush because the psyllid maximizes its reproduction and feeding activity on new leaves. Monitoring is especially important during this time and should include a search for all psyllid life stages, including adults, eggs and nymphs. Sampling methods vary in their effectiveness for different life stages, so an effective sampling program will utilize a combination of methods.

YELLOW STICKY TRAPS

Yellow sticky traps are used by the Citrus Pest and Disease Prevention Program (CPDPP) and California Department of Food and Agriculture (CDFA) to detect ACP adults for regulatory purposes, especially in regions not known to have the psyllid. They are most effective when hung at a one-



Photo 2: Visual examination of new citrus flush for evidence of Asian citrus psyllid adults, nymphs and eggs.

meter height on the outside rows of the orchard to capture ACP adults moving between orchard blocks. These traps are typically deployed at a density of at least one trap per half-mile of perimeter (one trap every 40 acres). They are collected and replaced every two weeks and are examined for the presence of adult ACP. If psyllids are found in a new area, a quarantine zone is established and trapping density is intensified.

The variation among individuals collecting samples is minimal for this method. However, yellow sticky traps are not as effective as other methods for quantifying ACP abundance because, when new flush is available, psyllids are more attracted to the volatiles and color of flush than yellow cards. This can lead to substantial underestimation of ACP populations at certain times of the year. Thus, yellow sticky cards are most effective when leaves have hardened off and when the psyllid population is high (**Photo 1**).

VISUAL INSPECTION

Visual inspection is one of the most important sampling methods, because it takes advantage of the adult psyllid preference for egg laying on new flush and the fact that immature stages develop only on young stems and leaves. Adult females deposit eggs on the tips of growing shoots or in the crevices of unfolded "feather flush" leaves. The eggs are tiny, almond-shaped, bright yellow-orange in color and are best observed by using a hand or head lens. The nymphs are found feeding on new flush and produce waxy tubules that direct honeydew away from their bodies. Sampling four new flushes per tree (when available) provides a reliable and consistent monitoring of all ACP stages. It is also important to record the stage of leaf as feather flush, growing flush or fully expanded leaves to determine the suitability of flush for nymphs.



Photo 3. Tap sampling for Asian citrus psyllid adults.

The visual search method is of limited use if there is no young flush available and individual differences in training among samplers may affect the consistency of this method (**Photo 2**).

TAP SAMPLING

The tap sampling method is common in Florida and Texas for monitoring adult ACP and is considered a rapid, reliable and consistent method. It can be used at any time of the year, regardless of the presence of new flush, because it collects adults from all types of foliage; but it is not helpful for monitoring psyllid eggs or nymphs, because they cling to the foliage. For tap sampling, a clipboard with plastic-coated white paper and a section of PVC pipe or similar device is needed. Adding a grid to the paper helps with counting ACP. A squirt of dish soap mixed in a half-liter of water is sprayed onto the clipboard to prevent psyllids from escaping before a count is made. The clipboard is held about one foot below a branch, and the branch is struck three times for each sample, causing the psyllids to fall on the clipboard. The number of psyllids on the clipboard are then counted and removed after each count, and the liquid is re-applied as needed. (Photo 3).

SWEEP NET SAMPLING

The sweep net method is also used for monitoring adult psyllids. As with tap sampling, it is not effective for monitoring eggs and nymphs. To perform this sampling method, a strong 15" diameter sweep net is required with a section of PVC pipe (optional). A branch (containing new flush, if available) is stuffed inside the rim of the sweep net and beaten with the PVC pipe three to four times consecutively for each sample, or the branch is vigorously shaken using the net. The adult psyllids on the foliage will fall inside the sweep net, and the collected psyllids are counted and recorded. If the number of ACP is low,



Photo 4. Sweep sampling for Asian citrus psyllid adults.

counting can be done by observing the contents of the sweep net carefully. If the number of ACP is high, the contents can be transferred to zip-lock bags or other containers and counted later. (**Photo 4**).

INTENT OF STUDY

The University of California recommended monitoring practice for visual, tap and sweep net sampling includes sampling at least 10 trees each on the border rows and in the center of the orchard for a total of 50 trees (see the UCIPM guidelines for citrus, Asian citrus psyllid recommendations: *http://ipm.ucdavis.edu/PMG/r107304411.html*). Sampling on multiple sides of trees or on alternating sides minimizes bias due to tree aspect. Research from Florida and Texas suggests that ACP tend to aggregate along the edges of the orchard and ACP densities are highest on the southeastern corners of orchards compared to other edges or the center of the grove. The purpose of our study was to examine the relative efficiency of tap and net sampling and to determine if there is any kind of edge effect in the distribution of adult psyllids under California conditions.

EVALUATING EFFICIENCY OF TAP VS. SWEEP NET SAMPLING AND EDGE EFFECTS

Twelve commercial southern California citrus orchards that included common citrus varieties (navel, grapefruit, lime and Valencia oranges) were sampled for adult ACP using the tap and sweep net methods from April to September 2014. These orchards were located in Riverside, San Diego and San Bernardino counties and varied in age, size and growth stages at the time of sampling. Each orchard was sampled once during the study. The orchards were not sprayed with pesticides for 30 days or more before the survey. In each site and for each method, four quadrants of each of 10 trees were sampled on the outside border of each of the north, east, south and west sides and a center row of the orchard. Hence, a total of 100 trees were sampled per orchard; 50 trees used for each sampling method. The total number of psyllids collected per tree (four sides combined) was summed and recorded in the field. Statistical analysis was performed using a linear mixed model in which the sampling method (tap or sweep) and sample location (N, S, E, W or Center) were defined as fixed effects, and the orchard was defined as a random effect.

The 12 orchards showed substantial variation in ACP population densities, with well over a 100-fold difference in mean ACP adult counts between the lowest (0.06 psyllids/tap or sweep) and highest density (19.48/sweep) sites (**Table 1**). The number of ACP adults collected per tree using a sweep net was higher than the number for tap sampling for 10 of the 12 orchards. In six of 12 cases, the difference was statistically significant, with 2.4 times more psyllids captured via a sweep net compared to tap sampling on average. At very low ACP densities (Sites 1 and 2), there were no differences in means. The variety of citrus did not seem to make a difference, affirming the literature that suggests that availability of flush is more important than citrus variety (Hall et al. 2008).

In contrast to the Florida and Texas experience, there were no significant differences in ACP numbers between the four sides or the center of the orchards (F 4, 24 = 0.04, P = 0.9963) for either sampling method (**Figure 1**). The number of ACP adults for the most different sides (North vs. South) were still within approximately ten percent of each other, and the center row didn't differ from the edge rows. Thus, ACP adults distributed themselves fairly evenly throughout these orchards.

Our results support the use of the sweep net sampling technique for monitoring adult ACP population because this method was consistently better able to capture adult psyllids compared to tap sampling, except at extremely low psyllid densities. The only limitation in the use of sweep net is that it is difficult to use on citrus varieties like limes and lemons because of their thorny nature.

The results of this study show that ACP adults were distributed evenly on the border rows, as well as in the center row. Earlier studies of within-field ACP distribution in commercial orchards in Florida and Texas found a significant "edge-effect" with more psyllids on the southeastern side (Setamou and Bartels 2015). This border effect has strongly influenced treatment strategies in Texas, where border treatments sometimes replace full field treatments. Interestingly, our findings suggest that no such affinity for specific edges of orchards exists in California and would indicate management strategies should be based on whole grove treatments. However, the current study was undertaken during the summer months when tender flush is low, and further research may find significant border effects at other times of the year, such as during the spring and fall flush.

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Table 1. Mean number of Asian citrus psyllid adults recorded using two methods in various locations.					
Site	Location	Variety	Mean no. of ACP/tree \pm S.D.		Fold difference
No.			Тар	Sweep net	
1	Temecula, Calif.	Navel	$0.06\pm0.24a$	$0.06\pm0.31a$	0
2	Highland, Calif.	Grapefruit	$0.16\pm0.50a$	$0.16 \pm 0.42a$	0
3	Redlands, Calif.	Navel	$0.38\pm0.67a$	$0.72 \pm 1.17a$	1.9
4	Redlands, Calif.	Navel	$0.42\pm0.64a$	$0.82 \pm 0.94b$	2.0
5	Riverside, Calif.	Navel	$0.44\pm0.76a$	$1.77 \pm 2.23b$	4.0
6	Highland, Calif.	Grapefruit	$0.77\pm2.49a$	$1.80 \pm 3.93a$	2.3
7	Redlands, Calif.	Navel	$0.78\pm2.35a$	$0.94 \pm 1.47a$	1.2
8	Mentone, Calif.	Valencia	$0.96 \pm 1.12a$	3.30 ± 2.64b	3.4
9	Riverside, Calif.	Navel	$1.02\pm1.58a$	2.51 ± 2.27b	2.5
10	Pauma Valley, Calif.	Lime	1.43 ± 1.99a	2.60 ± 2.56a	1.8
11	Redlands, Calif.	Navel	$3.74 \pm 3.21a$	7.30 ± 5.89b	2.0
12	Mentone, Calif.	Navel	$7.76\pm3.30a$	19.48 ± 10.23b	2.5
				Average difference	2.4

Means in each row followed by same letters are not significantly different (paired t-test, P < 0.05).

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