Sampling Methods as Abundance Indices for Adult *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in Citrus

H. N. NIGG,[1, 2] S. E. SIMPSON,[3] D. G. HALL,[4] L. E. RAMOS,[1] S. U. REHMAN,[1] B. BAS,[1] AND N. CUYLER[1]

Citrus Research and Education Center, Institute of Food and Agricultural Sciences, University of Florida, 700 Experiment Station Road, Lake Alfred, FL 33850

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ABSTRACT Beat sampling and two type of traps, cup traps and Tedders traps, were evaluated as sampling methods to detect and estimate population densities of adult *Diaprepes abbreviatus* L. weevils newly colonizing young citrus trees. The study was conducted over a 65-wk period across a 0.25-ha area of 80 citrus trees [*Citrus sinensis* (L.) Osbeck] (1.2–1.5 m tall). Beat samples were taken weekly to determine the number of trees infested and number of new adult weevils per tree. Sixteen of the 80 trees studied were each monitored weekly using one of the following trapping methods: cup traps in trees, cup traps on a stake in the ground within the tree drip line, cup traps on a stake in the ground outside of the drip line, Tedders traps on the ground within the drip line, and Tedders traps on the ground outside of the drip line. Weevils collected each week from trees and traps were removed from the study site. Based on the coefficients associated with Taylor's power law, the optimum numbers of trees to sample for an SEM equal to 25% of the mean estimate decreased from 50 trees at a mean of 0.5 new weevils per tree to 30 trees at a mean of 0.8 new weevils per tree. A significant relationship was found between the weekly mean number of new weevils per tree and the proportion of trees infested, a binomial relationship that could be further explored in the search for a sampling program for adult *D. abbreviatus.* Regression analyses indicated that three of the trapping methods served at least as weak indices of the presence and abundance of new weevils: cup traps in trees, Tedders traps inside the dripline and Tedders traps outside the dripline. Cup traps in trees and Tedders traps inside the dripline captured the most weevils and most frequently detected weevils. Although relatively inefficient as abundance indices of populations of new weevils, these two trapping methods appeared to have some value with respect to signaling when weevils first appeared in trees during the spring.

KEY WORDS *Diaprepes abbreviatus,* trap efficiency, root weevil, citrus, red navel

Diaprepes abbreviatus L. root weevil, is a polyphagous feeder (Fennah 1942, pp. 1–67) whose most economically important host in Florida is *Citrus* spp. (Simpson et al. 1996). Citrus is the most valuable agronomic crop in Florida; in 1998 it was produced on 378,182 ha, employed 89,700 people, and had an annual value of $9.13 billion (Hodges et al. 2002). Since its discovery in 1964, *D. abbreviatus* has spread to 20 counties in Florida and currently infests ≈66,420 ha (Anonymous 1997). *D. abbreviatus* may have been introduced to Florida on three occasions (Bas et al. 2000). The weevil is an important economic pest of citrus and difficult to manage (Hall et al. 2001). Certain management tactics can be implemented against egg-stage and adult-stage weevils, but these tactics need to be timed to coincide with the onset of large or peak densities of weevils in trees. This requires that a grower monitor for increases in population densities of adult weevils.

Sampling methods to monitor adult weevils in citrus are at an early stage of development. Various methods have been evaluated as surveillance tools for adult *D. abbreviatus* populations, although none appear to be effective (Beavers et al. 1979, 1982; Schroeder and Jones 1983, 1984; Jones and Schroeder 1984; Schroeder and Beavers 1985). Adult *D. abbreviatus* can be collected using traps such as cone-type emergence traps (Duncan et al. 2001), Tedders pyramid trap (Tedders and Wood 1994, Stansly et al. 1997, Duncan et al. 2001), and the Schroeder-Jones cup trap (Schroeder and Jones 1984). The Tedders trap is currently the trap recommended for monitoring for the presence of adult *D. abbreviatus* in citrus groves (Nigg et al. 2001). Cone-emergence traps provide information on absolute numbers of new adult weevils emerging from the soil, but only within the area under the trap and do not

[1] IFAS, Citrus Research and Education Center, University of Florida, 700 Experiment Station Road, Lake Alfred, FL 33850.

[2] E-mail: hnn@lal.ufl.edu.

[3] Florida Department of Agricultural and Consumer Service, 3027 Lake Alfred Road, Winter Haven, FL 33881.

[4] Research Department, United States Sugar Corporation, P.O. Box 1207, Clewiston, FL 33440.

provide information on numbers of immigrating weevils. Due to the small area that cone-emergence traps monitor, large numbers of traps are required for area-wide estimations of the number of emerging weevils. The capture potential of Tedders traps and cup traps (with respect to both newly emerged and older adults) are greater than cone-emergence traps, but the efficiency of these traps for detecting and estimating populations of the weevil has not been assessed.

Beat sampling, a common method of collecting insects (Borror et al. 1981), can be used to detect and collect *D. abbreviatus* weevils in citrus trees (Jones 1915, Nigg et al. 1999). In a previous experiment, a beat sampling procedure using umbrellas had an overall efficiency of 65% in estimating the absolute density of adult weevils in young trees and a detection efficiency of 75% (Nigg et al. 1999). Whether beat sampling could be used commercially to monitor adult weevils has not been investigated.

To time management tactics to coincide with peak adult weevil populations in trees, a sampling method that adequately measures number of weevils in trees and density fluctuations over time is needed. A sampling method effective at detecting new weevils emerging from soil and immigrating from other areas would be advantageous from the standpoint of timing management tactics early during a peak outbreak.

The purpose of this study was to compare beat sampling, cup traps and Tedders traps as sampling tools to detect and to provide indices of abundance of new adult weevils in young citrus trees.

Materials and Methods

In this study, beat sampling and five trapping methods were evaluated as methods of detecting and enumerating weevils newly colonizing young citrus trees. Counts from beat samples were used as an index of the absolute density of new weevils per tree. The relative efficiency of the trapping methods as indices of the presence and absolute density of new weevils per tree was evaluated and compared. A set of young citrus trees were sampled weekly, and weevils were removed from the trees as they were found. The study therefore focused on sampling for new weevils, either emerging from soil or immigrating from other trees.

Study Site and Experimental Design. The experiment was conducted in a 2.5 ha Red navel orange grove [*Citrus sinensis* (L.) Osbeck] planted in 1994 in Astatula sand with a 5–12% slope (Furman et al. 1975) at Mt. Dora, FL (274 trees/ha). Trees were 2.5 yr old (1.2–1.5 m tall) when the study was initiated and known to be infested by *D. abbreviatus* (Nigg et al. 2001). Beat samples were taken weekly for a 65-wk period (15 August 1996 – 6 November 1997) from each of 80 citrus trees to estimate number of trees infested and number of new weevils per tree. Each of 16 of the 80 trees was continuously monitored over the 65-wk period for adult weevils using one of the following trapping methods: (1) a cup trap on a stake in the ground midway between the dripline and trunk, (2) a cup trap on a stake in the ground 0.5 m outside the tree dripline, (3) a cup trap on a stake mounted in the tree, with the opening 1.0 cm above the canopy center peak, (4) a Tedders trap on the ground midway between the dripline and trunk, and (5) a Tedders trap on the ground 0.5 m outside the tree dripline. Traps were checked for adults once a week on the same day as beat samples were taken. The 80 trees sampled were subdivided into four plots of 20 trees, with each plot serving as a replicate. Each plot was five trees by four rows across, an area ≈15.4 by 24.6 m. Plots were separated by 10 trees within a row and by five rows (3 by 6-m tree spacing). Each trapping method was studied at four trees per plot, and the trees chosen for each trapping method were randomized within each plot.

The study site was a commercial and well-managed operation with an active pesticide program for the control of adult weevils. Records were maintained during the study on pesticide applications made for controlling adult weevils.

Beat Samples. Straight-handled golf umbrellas, 1.2 m diameter, were placed under the tree to cover the distance from trunk to dripline, and the foliage directly over the umbrella was beaten from top to bottom with an oak dowel (1.3 cm in diameter, 1.2 m in length). The umbrella then was moved to a new and contiguous area under the tree, the foliage was beaten, and this process was repeated until all of the foliage had been beaten. This method has been described by Nigg et al. (1999). Weevils were removed from the umbrella by hand, sexed (based on females and males having the tip of the abdomen pointed and rounded, respectively), and placed in a vial for removal from the field. Five people took data; four to beat and one to record data. Data were recorded as the number of weevils per tree. Weevils collected during beat sampling were removed from the grove to gain information on weekly number of new weevils in each tree, either newly emerged weevils or immigrants. To facilitate umbrella placement under the tree skirt, tree skirts were trimmed at the beginning of the study and periodically thereafter to provide a minimum of 0.3-m clearance between the tree skirt and the ground.

Sampling Statistics Associated with Beat Samples. The mean and variance were calculated each week for the number of weevils collected per tree by beat sampling ($n = 80$ trees/wk). The relationship between means and variances across the 65-wk study was investigated using Taylor's power law (Taylor 1961). The a and b parameters of Taylor's power law were estimated using a simple linear regression between $\log_{10}$-transformed means and variances for the weekly number of weevils per tree (Southwood 1978, Davis 1994). Optimum numbers of trees to beat sample to estimate mean population densities of new weevils per tree were estimated using the following equation:

$$n = (am^b)/(0.25\ m)^2,$$

where n = optimum number of trees to sample for a precision level of 0.25 (i.e., SEM 25% of the mean), a and b were the coefficients of Taylor's power law, and m was the mean number of weevils per tree

Table 1. Summary of trap and beat samples for new adult weevils

Trap	Mean (SEM) no. weevils per tree per week[a]		Mean (SEM) weekly % of trees identified as infested[b]	
	Trap samples	Beat samples	Trap samples	Beat samples
Cup trap inside drip-line	0.010 (0.003)	0.147 (0.015)	1.0 (0.3)	11.0 (1.4)
Cup trap outside drip-line	0.003 (0.002)	0.111 (0.012)	0.3 (0.2)	8.9 (1.2)
Cup trap in tree	0.051 (0.008)	0.160 (0.014)	4.3 (0.8)	12.8 (1.6)
Tedders inside drip-line	0.031 (0.005)	0.175 (0.016)	3.1 (0.6)	13.2 (1.8)
Tedders outside drip-line	0.012 (0.003)	0.163 (0.017)	1.2 (0.3)	11.9 (1.6)

[a] n = 1040 for each trap and companion beat samples.
[b] n = 65 for each trap and companion beat samples.

(Buntin 1994). A precision level of 25% was chosen because it enables the detection of a doubling or halving of a population (Southwood 1978). The relationship between mean weevil density per tree and proportion of trees infested was investigated by fitting weekly data to a mean-incidence model:

$$ln(m) = a' + b' ln\,(-ln[1 - p_T]),$$

where m was the mean number of weevils per tree, p_T was the proportion of trees infested, a' was the intercept, b' was the slope, and ln the natural logarithm (Jones 1994, Schaalje et al. 1991). The a' and b' parameters from the mean-incidence model were then used to estimate mean density per tree (m) based on p_T (Jones 1994):

$$m = \exp^{a'}[-ln(1 - p_T)]^{b'}$$

Trap Samples. The cup traps were a modification of traps described by Schroeder and Jones (1984). Our cup traps were constructed with a Solo 473 ml pale green plastic cup (R16 16 oz FF, Solo Cup, Urbana, IL) with the bottom removed and a cut-to-fit boll weevil trap top (Tedders and Wood 1994) pushed through the opening from the underside. The top was taped in place with duct tape. A 1.8-m bamboo garden stake was glued to a 1.3 by 1.3 by 0.5-cm piece of wood that was glued to the inside of the cup with construction adhesive (F-26, Leach Products, Hutchinson, KS). The small piece of wood allowed the stake to be placed so that it did not touch the inside of the cup, thus forcing a weevil crawling up the stake into the trap top. For the Solo cup trap mounted in the tree, bamboo stakes were cut to a length of 61 cm. The cup traps in trees were periodically adjusted to maintain the 1 cm position above the canopy. The modified Tedders traps were red and 61 cm tall (Tedders and Wood 1994). The hole in the screen cone of all the boll weevil trap tops was opened to 8–9 mm to accommodate larger adult *D. abbreviatus* weevils (Nigg et al. 1999). The trap top was determined to be 'escape proof' in a previous experiment (Nigg et al. 1999). The final data set consisted of 65 wk of beat counts for each tree and counts for each tree's companion trap.

Trap Efficiency. Trap efficiency was evaluated by comparing number of weevils captured in traps to number of new adults present in trees based on beat sampling, and by comparing percentage of trees identified as being infested by traps to percentage identified as infested by beat sampling. For each trapping method, regression analyses were conducted to determine if a significant relationship existed between mean number of weevils per trap and mean number of new weevils per tree based on beat samples (one analyses on raw data, one on $\log_{10}[x + 1]$). The fit of data to and slope of each regression were used to evaluate the relative efficiency of trap counts as an index of the absolute density of new weevils per tree. Regressions were also conducted between weekly percentage of traps with weevils and trees with new weevils ($\log_{10} x + 1$) to evaluate the trapping methods with respect to detecting trees infested by new weevils.

Results and Discussion

Adult *D. abbreviatus* were found on every tree beat-sampled during the study, and at least one infested tree was detected by beating during 53 of the 65-wk study. A weekly mean of 0.15 adults per tree (SEM = 0.01) was present over the 65-wk study based on beat samples. The largest number of weevils observed per tree was 10. A cumulative average of 9.80 weevils per tree (SEM = 0.69, range = 1–27, n = 80) was observed over the 65-wk study. Comparisons of the average number of weevils recovered and percentage of trees infested based on trap and beat samples are presented in Table 1.

Sampling Statistics Associated with Beat Samples. Over all weeks in which weevils were detected by beat sampling, regression analysis indicated that, for Taylor's power law, $a = 1.53$ and $b = 1.114$ [$\log_{10}$ (variety) = 0.18447 + 1.1137 $\log_{10}$ (mean); $F = 1098.9$, $P < 0.0001$, $r^2 = 0.96$, SEM of $a = 0.0338$, SEM of $b = 0.0336$, $n = 53$]. Optimum sample sizes ranged from ≈50 trees at a mean density of 0.5 weevils per tree to 30 trees at a mean density of 0.8 weevils per tree (Fig. 1).

Among the 53 wk during which new weevils were detected using beat samples, the percentage of trees infested each week ranged from 1.3 to 41.3% (mean = 14.2%, SEM = 1.33). The mean log-number of new weevils per tree (Y) was related to the proportion of trees infested (X): $ln(Y) = 0.2062 + 1.0437 ln(-ln[1-X])$; $F = 2386.7$, $P < 0.0001$, $r^2 = 0.98$, df = 52, b' standard error = 0.02136. Based on the parameters $a' = 0.2062$ and $b' = 1.0437$, the mean number of new weevils per tree (Y) was related to the proportion of trees infested (X) by: $Y =$

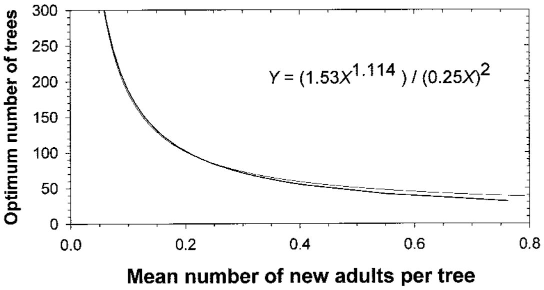

Fig. 1. Optimum (SEM 25% of mean estimate) number of young citrus trees to beat sample (*Y*) for an estimate of the number of new adult *D. abbreviatus* weevils per tree (*X*).

$\exp^{0.2062}[-ln(1-X)]^{1.0437}$ (Fig. 2). The results of these analyses may have limited value with respect to developing an applied binomial sampling plan for the weevil, because they were based on weekly counts of new weevils in trees and were obtained from a restricted range of means and percentages of infested trees, but the results were positive enough to indicate a binomial approach to estimating weevil densities deserves further investigation.

The *b* parameter (slope) of Taylor's power law (1.114, SEM 0.0336) was significantly >1.0 ($t = 3.274$, $a = 0.95$, df = 52), indicating that new adult weevils were aggregated among trees. This supported empirical observations, namely that a disproportionately large number of weevils were sometimes found within one or a few trees (e.g., see Harari and Landolt 1997). Whether removing weevils each week influenced the distribution of weevils among trees was unknown. Also, whether insecticides applied for control of *D. abbreviatus* or other pests influenced the distribution of adult weevils during the study was not known. Ten commercial pesticide treatments to control adult *D. abbreviatus* were made during the study: 15 August 1996, diflubenzuron (Micromite Uniroyal, Middlebury, CT) and petroleum spray oil (PSO, Exxon Mobile, Houston, TX); 28 October 1996, carbaryl (Sevin XLR, Aventis, Research Triangle Park, NC); 1 May 1997, Micromite, Sevin XLR and PSO; 8 May 1997, Micromite, Sevin XLR and PSO; 3 July 1997, abamectin (Agrimek, Syngenta, Greensboro, NC), Sevin XLR, Micromite, and PSO; 25 July 1997, Sevin XLR; 15 August 1997, formetanate hydrochloride (Carzol, Gowan, Yuma, AZ); 5 September 1997, Carzol; 29 September 1997, Sevin XLR; 6 October 1997, Sevin XLR. One week after each of four of these applications, adult weevils were not detected in any trees. However, after all eight applications, the number of new *D. abbreviatus* adults were as high or higher 2 wk after each application than just before each application. The reinfestation of trees by adult weevils was attributed primarily to continual emergence from the soil of new adult weevils.

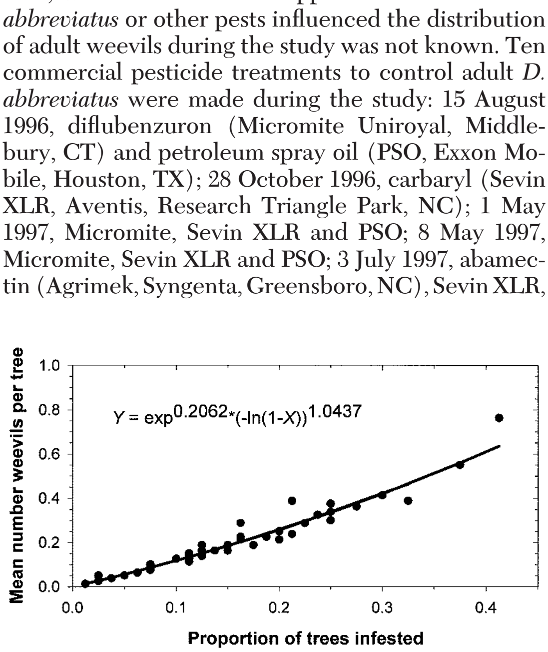

Fig. 2. Relationship between mean number of new weevils per tree (*Y*) and the weekly proportion of trees infested by new adult *D. abbreviatus* weevils (*X*).

Pesticide treatments may affect the behavior of an organism and the spatial pattern of a population of organisms in insecticide-treated versus and nontreated field plots (Tonhasca et al. 1994, Palumbo et al. 1995). Trumble (1985) observed changes in the dispersion pattern of *Tetranychus urticae* Koch in insecticide-treated strawberries, as surviving females may have migrated from treated areas and reproduced more rapidly. We do not know the effect of pesticide treatments on *D. abbreviatus* adult distribution or their influence on trap efficiency and relative captures, but each time a commercial pesticide application was made, our entire experimental area was treated. The spatial distribution of new adults and measures of trapping efficiency in our study should be valid for a grove under an intensive *D. abbreviatus* management program.

No data were taken on the time required to beat sample a young tree, but we estimated that perhaps 1 min would be required for trees 1.2–1.5 m tall. Beat sampling to obtain an estimate of the absolute number of weevils per tree might not be practical or possible in trees >2 m tall. However, a beating procedure to obtain a relative estimate of the population density of weevils in larger trees could be investigated.

Trap Efficiency. Cup traps on stakes in the ground inside of the drip line, cup traps on stakes in the ground outside the drip line, and cup traps in trees detected weevils during 9, 3, and 29 of the 65-wk study, respectively, (22, 7, and 67% of the week that new weevils were detected in trees, respectively). Tedders traps inside the drip line and outside the drip line detected weevils during 24 and 11 wk, respectively (52 and 26% of the week that new weevils were detected in trees, respectively). These data indicated that, during any given week, weevils were most likely to be detected in cup traps in trees or Tedders traps inside the drip line.

Regression analyses indicated that a significant relationship existed between the number of new weevils in trees and number captured in traps for three of the trapping methods: cup traps in trees, Tedders traps inside the dripline, and Tedders traps outside the dripline (Table 2). Correlation coefficients from these regressions indicated that estimates of weevil abundance in trees based on trap counts were weakly related to actual numbers of new weevils in trees. Among the three trapping methods, slopes associated with the regressions indicated weevil counts at cup traps in trees were most similar in magnitude to counts

Table 2. Comparison of weekly trap and beat samples for enumerating and detecting populations of new adult *D. abbreviatus* weevils

Sampling method	Fitted regression	*r*	*F*	*P*
Mean number of weevils per tree per week as indicated by trap (Y) or beat samples (X)[a]				
Cup traps inside dripline	$Y = 0.004–0.0004X$	0.00	0.00	(0.98)
Cup traps outside dripline	$Y = 0.001–0.0004X$	0.00	0.00	(0.98)
Cup traps in trees	$Y = 0.012 + 0.1331X$	0.26	4.45	(0.04)
Tedders inside dripline	$Y = 0.008 + 0.0728X$	0.28	5.49	(0.02)
Tedders outside dripline	$Y = 0.002 + 0.0457X$	0.27	4.92	(0.03)
Mean number of weevils per tree per week as indicated by trap (Y) or beat samples (X)				
Cup traps inside dripline	$Y = 0.010–0.0029X$	0.02	0.03	(0.87)
Cup traps outside dripline	$Y = 0.003–0.0012X$	0.01	0.01	(0.93)
Cup traps in trees	$Y = 0.032 + 0.1161X$	0.24	3.86	(0.05)
Tedders inside dripline	$Y = 0.021 + 0.0565X$	0.27	4.86	(0.03)
Tedders outside dripline	$Y = 0.006 + 0.0356X$	0.27	4.84	(0.03)
Mean percentage trees per week infested as indicated by trap (Y) or beat sample (X)[a]				
Cup traps inside dripline	$Y = 0.076–0.0619X$	0.12	0.95	(0.33)
Cup traps outside dripline	$Y = 0.019–0.0293X$	0.09	0.53	(0.47)
Cup traps in trees	$Y = 0.201 + 0.2886X$	0.36	9.51	(0.00)
Tedders inside dripline	$Y = 0.146 + 0.2466X$	0.33	7.61	(0.01)
Tedders outside dripline	$Y = 0.025 + 0.1577X$	0.29	5.84	(0.02)

[a] Analyses on $\log_{10}$ (data + 1). For all regressions, df = 64.

of new weevils in trees, but weevil counts at each of the traps were only a small fraction of the number of new weevils in trees. Each of the three trapping methods had some value with respect to identifying the percentage of trees that were infested by new weevils (Table 2). Correlation coefficients indicated that the traps were weak indices of the number of trees infested by new weevils, but somewhat better as indices of the number of new weevils per tree. Cup traps placed on stakes either inside or outside the tree dripline appeared to have no value as a sampling method for new adult weevils in trees.

Overall, cup traps in trees and Tedders traps inside the tree drip line generally provided data more similar to beat samples than other trapping methods. However, even these two trapping methods at a trap density of 16 per 80 trees were relatively inefficient as indicators of population densities of new adult *D. abbreviatus* in trees.

General Observations. Although the traps we tested were inefficient at the weevil densities in this study as tools to estimate population densities of new weevils in trees, and only moderately effective at detecting new adult weevils, cup traps in trees and Tedders traps within the drip line appeared to have at least some value in detecting when adult weevils first began increasing in trees during late spring and early summer 1997 (Fig. 3). For example, beat sampling indicated few adult weevils were present in trees during January–March 1997, but a noticeable increase in weevils was detected by beat sampling during mid-April 1997. Captures of weevils in cup traps in trees ranged from a mean of 0–0.05 per trap up until late April, and then the average number per trap increased to 0.4; this increase in the number of adults at cup traps, although about 2 wk later than indicated by beat samples, reflected the weevil increase based on beat sampling. Similarly, no weevils were captured at Tedders traps within the tree dripline during January–March 1997, but weevils were captured at these traps during mid-April at the same time beat samples indicated weevil populations were increasing.

In total, 435 female and 344 male weevils were observed during beat sampling over the 65-wk study. More females than males were generally observed during fall 1996 (69 females, 38 males) and fall 1997 (84 females, 44 males) than during other seasons of the year.

We conclude that the best placement for the Tedders trap is under the dripline and for the cup trap in the tree. However, the use of either of these traps for monitoring the presence or abundance of *D. abbreviatus* would be tenuous. Researchers have long been interested in bait trapping to monitor adult weevils, but, despite serious efforts in this direction, no suitable attractants have been found (Beavers et al. 1982, Schroeder and Jones 1983, Jones and Schroeder 1984, Harari and Landolt 1997). It remains possible that other sampling methods might exist for monitoring

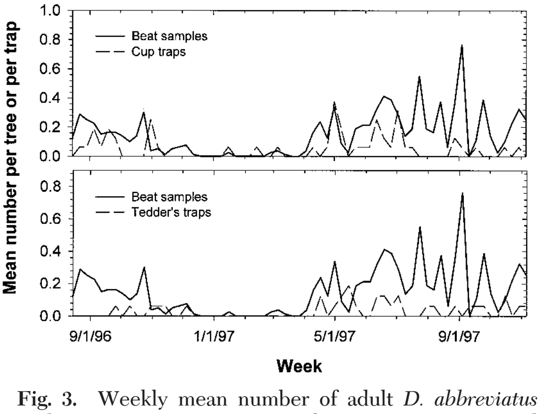

Fig. 3. Weekly mean number of adult *D. abbreviatus* weevils per trap (16 traps per week, cup traps in trees and Tedders traps inside the drip line) and weekly mean number of new weevils per tree based on beat samples in young citrus trees (80 trees beat-sampled per week).

population levels of adult weevils, i.e., relating number of flush shoots with fresh weevil feeding damage to population densities of adult weevils.

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