

Spatial Dispersion and Binomial Sequential Sampling for Citricola Scale (Homoptera: Coccidae) on Citrus

J. T. TRUMBLE, E. E. GRAFTON-CARDWELL, AND M. J. BREWER²

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ABSTRACT A binomial (presence-absence) sequential sampling plan for citricola scale, *Coccus pseudomagnoliarum* (Kuwana), was developed and validated for infestations on orange trees. Population densities from 200 collections of 25 leaves in both 1991 and 1992 were used to determine the Green coefficient (C_x), and the intercept and slope values for both the Taylor power law and Iwao regression of mean crowding on the mean. All measures of dispersion agreed in both years that scale populations were aggregated; C_x was > 0.3 and slopes from the regressions were significantly > 1 ($P < 0.001$). Because the fit of the regression lines was best using the Taylor power law, a binomial sampling plan was developed incorporating power law coefficients into Wald's (1947) approach with upper and lower thresholds, a critical action level of 1 scale per leaf, and a truncation at 200 samples. A minimum sample of 25 leaves collected from different trees is recommended to minimize potential incorrect treatment decisions caused by among-tree variation. In 11 validation trials in 1993, the binomial plan compared favorably with a conventional technique where citricola scales on 125 leaves from 5 trees were counted. Both techniques reached the same conclusions regarding the need for treatment in 10 of 11 trials, but the binomial plan required less average time while allowing more trees to be examined.

KEY WORDS binomial sampling, presence-absence, sequential sampling

IN THE EARLY 1900s, the citricola scale, *Coccus pseudomagnoliarum* (Kuwana), was one of the most damaging insects on citrus in the San Joaquin Valley of California (Quayle 1938, Kennett 1988). Damage occurred in the spring before harvest when the overwintering scales began to grow rapidly and produce honeydew, reducing fruit production and resulting in sooty mold growth on the remaining fruit (Quayle 1915). This insect continued to have sporadic outbreaks but was largely eliminated as a pest in the 1940s by the advent and use of DDT and subsequent organochemicals (Elmer et al. 1980). However, as these compounds lose effectiveness because of resistance development, and consumer acceptance of pesticide residues decrease, growers are gradually turning to the use of selective pesticides and natural enemies for citrus insect control; including botanicals such as sabadilla for citrus thrips control and microbials for lepidopterous pests. These pesticides are ineffective on citricola scale. This has led to concerns that resurgence in citricola scale populations could occur within 4-5 yr of the elimination of broad spectrum pesticides (Elmer et al. 1980, Kennett 1988, Georghiou and Lagunes-Tejeda 1991). Such resurgences are now occurring in orchards in the San

Joaquin Valley (E.E.G.-C., personal observation) and have been reported in the desert areas of Arizona (Bartlett 1978).

An efficient and statistically accurate sampling plan for citricola scale is needed to assess population changes on oranges. This insect appears to be a good candidate for sequential sampling because it is univoltine, the crawler stage is short (2 d), and cohorts are closely synchronized following egg hatch in early summer (Quayle 1915, Gressitt et al. 1954). Thus, populations need to be sampled only once each year per location. Because the insect causes little direct feeding damage in the summer, and management practices timed for September or October provide good control (Elmer et al. 1980), a substantial window of time exists at each location in which to sample without concern for economic loss. In addition, summer populations are located almost entirely on the underside of the leaves (Quayle 1915), and this plant part provides an easily sampled component which, because of the low mobility of the insect, represents the "natural habitat unit" for sampling as defined by Patil and Stiteler (1974). Therefore, the primary objectives of this study were to document the spatial dispersion pattern of the citricola scale, and then use this information to develop and validate a sampling plan suitable for rapidly estimating population densities during August through October.

¹ Department of Entomology, University of California, Riverside, CA 92521.

² Department of Plant, Soil and Insect Sciences, University of Wyoming, P.O. Box 3354, Laramie, WY 82071.

Materials and Methods

Citricola Scale Distributions. Numbers of immature citricola scales per leaf were counted on samples of 25 leaves collected from each of 200 trees in both 1991 and 1992 in a 4.05-ha block of Valencia oranges in Tulare County, California. Leaves were arbitrarily collected from the lower half of the northeastern most quadrant of the tree, because previous reports indicated that infestations were most commonly found in this area (Quayle 1915, Elmer et al. 1980). The population densities on these leaves were used to calculate three commonly used dispersion indices, including the Green (1966) coefficient C_x , Iwao (1968, 1970) regression of mean crowding on the mean, and Taylor power law (Taylor 1965). Three such indices were chosen in an attempt to get a consensus on dispersion, because use of a single index can be misleading (Myers 1978). C_x was determined using the formula (Green 1966):

$$C_x = \frac{[s^2/m] - 1}{\Sigma x - 1}$$

where s^2 = variance of the mean, m = mean number of citricola scale in i sampling units, and Σx = total number of citricola scale sampled in i sampling units. The regression method of Iwao was calculated by solving the equation $m = \alpha + \beta m$, where α (estimated by a) = the intercept on the ordinate, and β (estimated by b) = the slope of the regression line when m is regressed on the mean. Mean crowding, m , was derived from the Lloyd (1967) formula $m = m + (\sigma^2/m - 1)$, and substituting the mean and variance from the count data. Student t -tests were used to determine if $b = 1$. Regressions and t -tests were generated using Statview and SuperAnova (Abacus Concepts 1991).

The coefficients a and b of the Taylor power law, $s^2 = a m^b$, were estimated from the regression equation: $\log(s^2) = \log(a) + (b) \log(m)$, where a and b = the intercept and the slope coefficients, respectively. Regressions and t -tests were generated using Statview and SuperAnova (Abacus Concepts 1991).

To determine if the leaf samples we collected, as well as the conventional scouting technique of sampling 125 leaves from 5 trees, were estimating population densities with a reasonable level of precision, the Green (1970) formula for estimating mean density at specific precision levels was calculated. This was accomplished at precision levels (Do) of 0.25 and 0.20 by solving:

$$\log T_n = \frac{\log(Do^2/a)}{b-2} + \frac{b-1}{b-2} \log n$$

where T_n = the cumulative number of citricola scale counted, a and b were the intercept and slope parameters of the Taylor power law, and n = the number of leaves collected.

Development of the Binomial Sequential Sampling Plan. The Wald sequential sampling

plan was constructed using safe and action thresholds of 0.5 and 1.0 citricola scale per leaf, respectively (after Brewer et al. 1994). The safe threshold was defined as the proportion of infested sample leaves at or below which there is a reasonable expectation that a management action would not be economical. The action threshold was that proportion of infested leaves at or above which there is good confidence that a management action would be economically justified. The action threshold of 1.0 scale per leaf was selected because this is the current level accepted and used by pest-control advisors in San Joaquin Valley citrus (E.E.C.-C., personal observation). The safe threshold of 0.5 citricola scale per leaf was selected because it is a level which pest-control advisors do not consider harmful (J. Stewart, personal communication). Based on the Wald (1947) argument, making a decision when the infestation rate is between the safe and action thresholds is of little practical importance. Decisions were designed not to exceed a nominal error rate of 0.10 (from Wald [1947], $\alpha = \beta = 0.01$).

The action and safe thresholds corresponded to proportion values of 0.57 and 0.41, respectively, to be used in the binomial sequential sampling plan. This correspondence was obtained by applying the average coefficient values from the Taylor power law (differences were not significant between 1991 and 1992 data at the $P < 0.05$ level, F test [Sokal and Rohlf 1969]) to the following equation from Wilson and Room (1983):

$$P(I) = 1 - e^{-\frac{x[\ln(ax^{b-1})]}{[ax^{b-1}] - 1}}$$

where $P(I)$ = the proportion of infested leaves, a = the intercept from Taylor power law, b = the slope from Taylor power law, and x = the density of scale per leaf. The safe and action threshold proportions and error rates of 0.10 were fit to Wald (1947) equations to produce the upper and lower decision threshold lines of the sequential plan. Although previous reports indicated that distributions were uniform throughout an area of infestation (Bartlett 1953), our preliminary data indicated some among tree variation, and we chose to take the conservative approach that a minimum of 25 leaves from each sampling site (1 leaf per tree) would be collected.

Sampling was set to terminate at 200 leaves if a decision was not made by this time. At this time, the midpoint of the safe and action thresholds can be used as a single decision threshold for making a plan-terminating decision. Inspection of the average sample number curve will assist in determining if the truncation mark of 200 results in a desirable low frequency of truncations of the plan. Simulations can also be constructed to determine the maximal frequency of the occurrence of plan truncation.

Table 1. Dispersion indices for citricola scale in 1991 and 1992

Year	Range of means ^a	Taylor power law			Iwao regression			Green index
		a	b	r ²	a	b	r ²	
1991	0.1-3.1	1.23	1.64	0.917	-0.34	4.22	0.605	0.310
1992	1.1-11.3	1.49	1.57	0.842	5.15	2.12	0.651	0.301

Taylor power law and Iwao regression were significant at the $P < 0.001$ level.

^a Mean citricola scale per leaf for 200 samples per year of 25 leaves each.

The average sample number and operating characteristic curves for the binomial sequential sampling plan were calculated using simulations of presence-absence data as reported by Brewer et al. (1994). For these simulations, the proportion of infested samples was varied from 0.01 to 0.99 in increments of 0.01 or in increments of 0.005 when in the vicinity of decision thresholds. The simulations were run 1,000 times for each incremental infestation. Average sample number values were generated when each simulation was terminated by plan specifications by dividing the cumulative number of infested samples by the cumulative number of outputs, each summed across 1,000 runs. Operating characteristic probabilities were calculated as the proportion of the 1,000 runs in which the decision was made that the proportion of infested samples was at or below the safe threshold. To determine the percentage of sampling episodes that would not result in a recommendation during the sequential phase of the plan, these simulation runs were partitioned into those that resulted in either *treatment necessary* or *no treatment necessary* conclusions before sampling 200 units and those that resulted in making a plan-terminating decision once 200 leaves were inspected.

Validation of the Sampling Plan. Validation of the sampling plan was conducted at 11 locations in 1 Valencia and 2 navel orchards in Tulare and Madera counties in September and early October of 1993. At each location, comparisons were made between the conventional scouting technique of 125 leaves from 5 trees and the binomial sequential sampling plan. A minimum of 25 leaves per site (1 per tree) was collected for the sequential sampling approach. Surveying >5 trees appeared advisable because scouts tend to examine many trees in an area to ensure that no unusual or unexpected problems have developed since the last evaluation was conducted. On 4 occasions, the time necessary to complete each sampling technique was recorded.

Results and Discussion

Citricola Scale Distributions. Mean scale densities per leaf were generally low in 1991, but ranged up to 3-fold the action threshold of 1.0 citricola scale per leaf (Table 1). In 1992, populations were higher, sometimes exceeding the critical treatment level by >10-fold. Thus, the range of scale densities from samples collected in these 2

yr spans most of the densities likely to be encountered by scouts in commercial production orchards, as well as the safe and action thresholds of the proposed sampling plan.

All indices were in agreement that the populations of citricola scale were aggregated (Table 1). Green coefficient was >0, indicating the population was aggregated rather than random. In both 1991 and 1992, slopes of the regression lines for the Taylor power law were quite similar and not significantly different ($P > 0.05$). Although the fit of the regression was generally poorer with the Iwao regression (see Table 1), the slopes in both years were also significantly >1, thereby indicating scale populations were aggregated (1991, $t = 25.22$, $P < 0.001$; 1992, $t = 30.47$, $P < 0.001$).

Green (1970) fixed precision level sampling curves for both 1991 and 1992 were quite similar, rarely diverging by >10 insects for any given number of leaves sampled (Fig. 1). The slopes of these lines began to flatten substantially between 50 and 60 leaves sampled, particularly at the precision level of 25%. This information suggests that the conventional scouting technique of counting 125 leaves per sampling stop should provide a mean density estimate with considerable accuracy, even at relatively low population densities. Thus, comparing any proposed binomial sampling approach with the robust conventional counting technique currently used for citricola scale would provide a stringent validation test for the binomial strategy.

Development of Binomial Sequential Sampling Plan. The binomial sampling plan for citricola scale presented in Fig. 2 is used by collecting 1 leaf per tree from the northeast quadrant and noting the presence or absence of citricola scale. As sampling proceeds, if the cumulative number of infested leaves is less than the lower decision threshold line, then sampling can be stopped and no suppression treatment is required. If the cumulative number of infested leaves is above the upper decision threshold, then the action threshold has been exceeded and a management action is warranted. If the cumulative number of infested leaves falls between the upper and lower thresholds, additional leaves are sampled to a maximum of 200. In the unlikely event that 200 leaves are sampled without a treatment decision being reached (see Fig. 3a), the sample proportion can be compared with the midpoint of 0.57 and 0.41 proportion of infested leaves (=98.6 infested leaves) of a 200 leaf sample. Thus, if ≤ 98 leaves

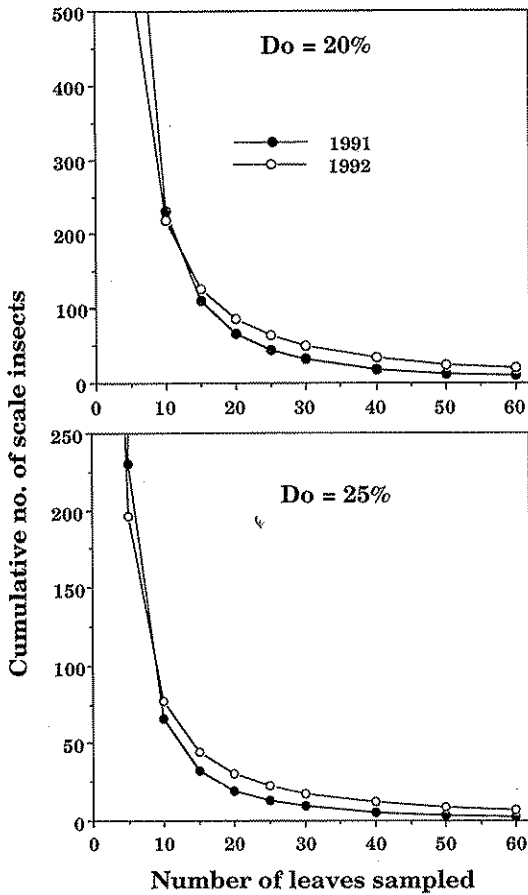


Fig. 1. Cumulative numbers of citricola scale needed for variable numbers of samples to estimate population density with fixed precision levels of 20% and 25%.

have citricola scale, a suppression treatment is not warranted; if ≥ 99 leaves have scale present, then a management action is justified. A minimum sample of 25 leaves (at one leaf per tree) is recommended to minimize potential incorrect treatment decisions caused by among-tree variation.

The average sample number curve indicated that the maximal average sample number for the binomial sampling plan would be 52 (Fig. 3a). This value takes into consideration that 1.5% of the time a plan-terminating decision would be made only after 200 leaves were inspected. This maximal average sample number occurred at the midpoint of the upper and lower threshold proportions of 0.57 and 0.41, respectively. Therefore, the greatest amount of sampling effort would be expected to occur at infestations where $\approx 50\%$ of the leaves are infested. The minimal average sample number was 7, but would not be used if a 25-leaf minimum were employed.

Despite the use of a truncation mark, which was incorporated into the simulation, the operating characteristic curve had an acceptable shape (Fig.

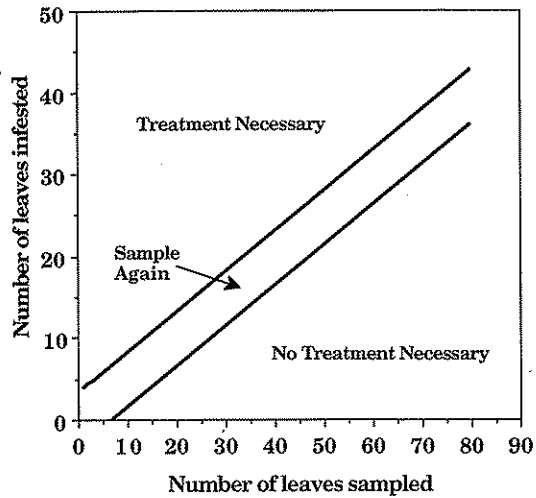


Fig. 2. Binomial sequential sampling plan for citricola scale covering the sample sizes encountered in the validation trials in 1993. In practice, presence or absence of scales was recorded for leaf samples, and then plotted against the number of leaves collected. Sampling continued until a treatment decision was reached or 200 leaves were sampled.

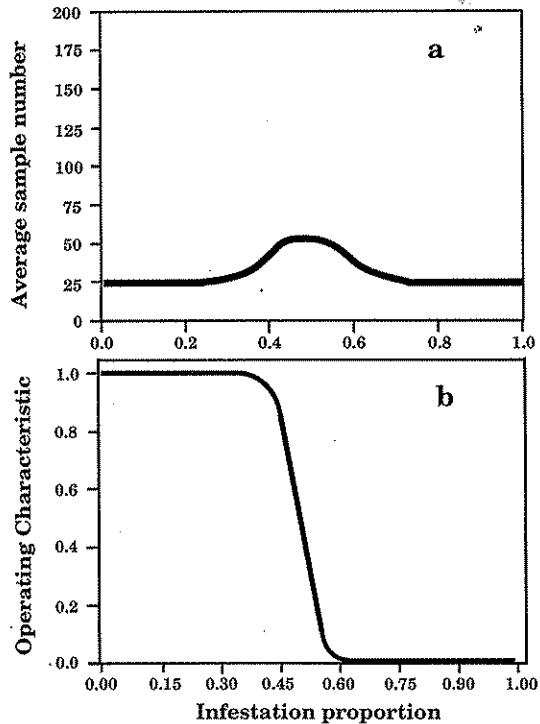


Fig. 3. Average sample number (a) and operating characteristic curves (b) for the binomial sequential sampling program for citricola scale on orange tree leaves. Decision thresholds are described in the text; truncation was set at 200 leaf samples.

Table 2. Validation of the binomial sequential sampling plan in 1993 by comparison with conventional sampling.

Sample location	Conventional sampling		Binomial sequential sampling		
	Mean/leaf	Control needed	No. leaves sampled	No. infested	Control needed
1	1.72	Yes	25	18	Yes
2	1.34	Yes	25	20	Yes
3	3.50	Yes	25	18	Yes
4	4.10	Yes	25	21	Yes
5	0.69	No	25	4	No
6	5.67	Yes	25	16	Yes
7	3.78	Yes	85	44	—
8	0.05	No	25	1	No
9	0.03	No	25	0	No
10	0.01	No	25	0	No
11	0.10	No	25	0	No

—, Sampling prematurely terminated before a decision was reached.

3b). Brewer et al. (1994) showed that operating characteristic curves with truncation marks of 200 did not differ substantially from those performing without truncation marks under commonly used upper and lower threshold conditions.

Because this plan uses a truncation mark, there will be times when the plan is terminated once 200 leaves are sampled (terminal decision) instead of during the operation of the sequential part of the plan (sequential decision). Under the simulation conditions of Brewer et al. (1994), the maximal frequency of terminal decisions occurred at the midpoint of the upper and lower threshold proportions of 0.57 and 0.41, respectively. For our data, simulation results indicated that the frequency of terminal decisions average 1.5% of the judgments made in 1,000 simulations.

Validation of Sampling Plan. The binomial sampling plan compared favorably with the conventional scouting plan (Table 2). In 10 of 11 comparative trials in 3 orchards in September and October 1993, both approaches reached the same conclusions in all instances (Table 2). This agreement occurred not only for extremely low or high densities, but also for scale densities close to the critical treatment level of one scale per leaf. Thus, the resolving power of the binomial sampling plan at critical densities appears at least equivalent to the conventional technique. In one trial where no decision was reached, sampling was terminated prematurely (mistakenly) when the binomial plan required the same amount of time as the conventional scouting plan.

The time required for sampling was substantially reduced when the binomial plan was employed. In 10 of the 11 trials, only 25 leaves (our minimum sample size) needed to be examined for presence of the scale before a decision was reached using the binomial plan. The average time required to examine 25 leaves was 5 min 20 s. This represents a substantial reduction from the 17 min 35 s re-

Table 3. Example citricola scale sampling sheet

Sample size, leaves	No. infested leaves	Below	Above	In between?
		no., no treatment	no., apply treatment	
25	—	9.0	15.7	Continue sampling
40	—	16.4	23.0	Continue sampling
55	—	23.8	30.4	Continue sampling
70	—	31.2	37.8	Continue sampling
85	—	38.6	45.2	Continue sampling
100	—	46.0	52.6	Continue sampling
115	—	53.4	60.0	Continue sampling
130	—	60.8	67.4	Continue sampling
145	—	68.2	74.8	Continue sampling
160	—	75.6	82.2	Continue sampling
175	—	83.0	89.6	Continue sampling
190	—	90.3	97.0	Continue sampling
200	—	95.3	101.9	Stop, no decision

quired for the conventional counting technique. In the one trial where >25 leaves were required before a decision was reached, 85 leaves were examined. Not surprisingly, the additional sampling effort was required when scale density was close to the critical treatment level. The time required to examine a leaf from 85 different trees was equivalent to that required to sample and count 125 leaves from 5 trees. However, the time advantage of the binomial sampling plan could be further improved by sampling 5–15 leaves per tree if >25 leaves need to be sampled, thus reducing transit time between trees while maximizing the number of trees visited. An example data sheet for the binomial sequential sampling plan for citricola scale using the aforementioned strategy has been presented in Table 3. The final line indicates no decision if counts of 200 leaves produce a cumulative number of infested leaves between 93.5 and 101.9. However, a conclusion with likelihood of error of $\approx 10\%$, the same as when a sequential decision is made, could be reached if the midpoint of 98.6 infested leaves were chosen as a single threshold; above this value treatment is applied, below this value no management practice is necessary.

No sampling plan should be assumed to work for every occurrence of an insect. Geographic variation in growing conditions and changes in pesticide use patterns between locations have been proven to be significant sources of variability in accuracy and effectiveness of sequential sampling plans (Trumble 1985, Trumble et al. 1989). In particular, the sampling program presented for citricola scale should be validated for other types of citrus before adoption on those crops. However, the binomial sequential sampling technique is rapid enough that comparisons with conventional sampling programs could be conducted with a minimum of effort.

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