Spatial Distribution of Adult *Bemisia tabaci* (Homoptera: Aleyrodidae) in Cotton and Development and Validation of Fixed-Precision Sampling Plans for Estimating Population Density

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ABSTRACT We conducted studies to examine distributional patterns of adult *Bemisia tabaci* (Gennadius) strain B (also referred to as *Bemisia argentifolii* Bellows & Perring) in cotton, *Gossypium hirsutum* L., and to develop and validate a sequential sampling plan for estimating population density. Adults were consistently more abundant on mainstem leaves from the top stratum of cotton plants than on mainstem leaves from the middle and bottom strata. Counts on mainstem leaves from the top of the plant also had the lowest relative variation. Adults on the top stratum of the plant were fairly uniformly distributed over leaves from mainstem nodes 2–7 (terminal = node 1), but numbers of adults were highest and least variable on fifth-node leaves. Patterns of aggregation, as measured by Taylor's power law, did not differ among the top, middle, and bottom strata of cotton plants and were similar among the first six mainstem leaves below the mainstem terminal. Ratios between counts of adults on individual leaves from the top stratum of the plant and whole plant counts were variable and averaged (±SD) 0.075 ± 0.071. Based on fifth mainstem node leaves as the sample unit, we used Kuno's and Green's methods to develop fixed-precision sequential sampling plans. The underlying mean–variance models for these methods and performance of the sequential stop lines were compared and evaluated using a resampling simulation of independent data sets with means ranging from 2 to 50 adults per leaf. Compared with Iwao's mean crowding regression, Taylor's power law was a less biased predictor of variance. As a result, Green's plan, on average, achieved the desired precision better than Kuno's plan even though neither plan consistently gave mean estimates with the desired precision. Further simulations provided preliminary adjustments in the stop lines for field implementation.

KEY WORDS *Bemisia tabaci* strain B, spatial distribution, sampling plans

THE SWEETPOTATO WHITEFLY, *Bemisia tabaci* (Gennadius) strain B (also referred to as *B. argentifolii* Bellows & Perring), continues to be a devastating pest of cotton and several spring and fall vegetables in Arizona and southern California. Populations of this insect develop continually in the southwestern United States, sequentially moving among various wild host plants and cultivated crops throughout the year (Natwick & Zalom 1984, Butler & Henneberry 1986, Coudriet et al. 1986, Watson et al. 1992, Norman et al. 1993). The polyphagous habit and interhost movement of this insect contributes to the complexity and difficulty of pest management. In particular, efficient monitoring and management of this pest over a large area and in a number of cropping systems is required.

Reliable and cost-effective sampling methods are central to study of the biology and ecology of *B. tabaci* and are critical to the development of monitoring programs for pest management application. Recently, several studies have examined the spatial distribution of *B. tabaci* immature stages on several major crops, including peanuts (Lynch & Simmons 1993, McAuslane et al. 1993), cantaloupe (Tonhasca et al. 1994), and cotton (Naranjo & Flint 1994). Several of these studies have also formulated sampling plans for the efficient estimation of eggs and nymphal population density (Naranjo & Flint 1994, Tonhasca et al. 1994). These methods permit efficient monitoring of eggs and nymphs for research purposes, but because it is difficult to make accurate counts of these stages in the field it is unlikely that these sampling plans will be useful for pest management application. In contrast, adults are relatively easy to monitor, and a variety of methods have been developed and used worldwide for sampling this nonsedentary stage (see Butler et al. 1986, Ohnesorge & Rapp 1986, Ekbom & Xu 1990).

There has been limited effort to describe the spatial distribution of adult *B. tabaci* (Butter & Vir 1990, Rao et al. 1991, Naik & Lingappa 1992, Liu et al. 1993, Tonhasca et al. 1994) or develop formal

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sampling plans (Tonhasca et al. 1994). Here we describe the within-plant and within-field distribution patterns of adult *B. tabaci* in cotton in Arizona, define an efficient sample unit, relate counts on this sample unit to absolute density, and present a sequential sampling plan that allows the estimation of adult density at fixed levels of precision. Further, we validate the performance of our sampling plan against independent field data using a resampling simulation method and suggest preliminary refinements to the plan for field implementation.

Materials and Methods

Within-Plant Distributions. We examined within-plant distributions of adult *B. tabaci* on seven occasions in 1992 and 1993. In 1992, samples were collected from 24 plots of two upland cottons, *Gossypium hirsutum* L., Deltapine 50 ('DP-50') and Stoneville 506 ('ST-506') at the University of Arizona, Maricopa Agricultural Center (MAC), Maricopa, AZ. These plots were part of a split-plot design experiment to evaluate the effect of irrigation scheduling on *B. tabaci* population dynamics (Flint et al. 1994a). The 24 individual plots (≈0.1 ha) were planted on 17 April and were arranged contiguously in a 2.4-ha area with 2-m borders between plots. On each of four dates (16 June and 1, 16, and 29 July) we counted the number of adults on the undersides of mainstem leaves from the top, middle, and bottom thirds of the plant. Hereafter these are denoted as top, middle, and bottom strata. Over that period of time the average (±SEM, $n = 12$) number of mainstem nodes per plant increased from 13.9 ± 0.7 to 21.5 ± 2.3. Counts were made by carefully turning the leaf over by rotating the petiole or the tip of the leaf blade. To reduce disturbances that might have interfered with an accurate census, counts were done on a single leaf (top, middle, or bottom) on each of three consecutive plants at a sample site. These counts were done at five randomly selected sites per plot in each of 24 plots (12 plots per cultivar). In 1993 we collected samples on 30 July and 12 August from a 0.4-ha field of upland cotton (Deltapine 90 ['DP-90']) adjacent to the USDA–ARS Western Cotton Research Laboratory, Phoenix, AZ, and from four biweekly irrigated 0.1-ha plots of 'DP-50' at the MAC. Because results from 1992 indicated that adults were most abundant in the top stratum of the plant, we counted adults on individual mainstem leaves from nodes 2 through 7 (terminal = node 1). Similar to 1992 procedures, we counted adults on a single leaf on each of six consecutive plants at a sample site. Counts were taken from 60-90 randomly selected sites per sampling date. Plant densities averaged 65,000/ha with 1.02-m row widths in all fields in both years. All counts were completed between 0600 and 0800 hours. In 1992 and 1993 the average minimum/maximum temperatures during sampling dates over this interval were 18/26°C and 19/23°C, respectively.

A mixed-model analysis of variance (SAS Institute 1988) was used to test for differences in abundance relative to vertical strata (1992) or nodal position within the plant (1993), date of sampling, cultivar, and the interactions of these factors. Initially, irrigation regime was entered as an additional factor in the 1992 analysis. Neither irrigation nor any of the interaction terms including irrigation were significant ($P > 0.1$), and so this factor was removed in the final analysis. The elements plants-within-dates (1993) or plants-within-date by cultivar (1992) were considered random factors. All counts were transformed by $\ln(x + 1)$ before analyses. Because of low counts of adults on the first two dates in 1992, only the final two dates were included in the analysis for that year.

To test for differences in levels of aggregation between strata in 1992, we used Taylor's power law (Taylor 1961), $S^2 = am^b$, where S^2 is variance, m is mean density per leaf, and a and b are fitted parameters. We regressed $\ln(S^2)$ on $\ln(m)$ of counts from each stratum and used analysis of covariance (SAS Institute 1988) to test for heterogeneity of regression parameters. A similar analysis to compare levels of aggregation between leaves from nodes 2–7 was not performed in 1993, because only three data points were available for counts from each node.

Estimation of Absolute Density. Concurrent with individual leaf samples, we counted adult *B. tabaci* on whole cotton plants on six dates. In 1992 we sampled two randomly selected plants per plot, with 24, 10, 12, and 10 plots being sampled on 1, 16, and 29 July, and 12 August, respectively. In 1993 we sampled 20 plants on each of two dates (30 July and 12 August). We captured adults using muslin sleeve cages (1.5-m height by 0.5-m diameter) fitted with drawstrings on either end. Forty-eight hours before each sampling date, these cages were placed around the base of plants, and one drawstring was tightened around the stem to seal the cage just above soil level. On the day of sampling the bag was carefully pulled up over the plant, and the top drawstring was tightened. The stems were cut at ground level, and the bagged plants were brought to the laboratory and chilled for 1–2 h at 4°C. After chilling, the plants and the inside surface of the cages were then searched for *B. tabaci* adults. All samples were collected between 0600 and 0800 hours.

We averaged the individual leaf counts (top stratum or fifth mainstem node leaf) and the individual whole plant counts for each plot on each sample date. We calculated the ratio between these means and also performed regression analysis to relate leaf and whole plant counts. We examined differences between cultivars in 1992 by using analysis of covariance (SAS Institute 1988) to test for heterogeneity in regression parameters.

Within-Field Distributions. Samples were collected weekly to biweekly on 16 dates from 8 June through 30 September 1993 from upland ('DP-50') and long-staple, *G. barbadense* L. ('Pima S-7') cotton arranged in 24 0.1-ha plots (12 per cultivar) at the MAC. Again, plots were arranged using a split-plot design in a contiguous 2.4-ha area (≈65,000 plants per ha, 1.02-m row widths) with half the plots receiving weekly irrigation and half receiving biweekly irrigation (Flint et al. 1994b). Adult *B. tabaci* were counted on the underside of mainstem node leaves from the fifth node (see results for definition of the sample unit) on 15 randomly selected plants per plot. This resulted in 64 treatment/date combinations with 90 individual leaf counts per observation for calculating means and variances. All counts were completed between 0600 and 1000 hours. The average minimum and maximum temperatures during sampling dates over this time interval were 19 and 29°C, respectively.

Sample Plan Development. Fifty-nine of the 64 observations above had means >0 and were usable for estimating parameters for two mean–variance models: Taylor's power law and Lloyd's (1967) mean crowding index, $\overset{*}{m} = m + (S^2/m - 1)$. For Taylor's power law we regressed $\ln(S^2)$ on $\ln(m)$ to derive estimates of a and b, and for mean crowding we regressed $\overset{*}{m}$ on m to derive estimates of α and β from $\overset{*}{m} = \alpha + \beta m$ (Iwao 1968, 1977). For the mean crowding model the variance is related to the mean as $S^2 = (\alpha + 1)m + (\beta - 1)m^2$. Mean–variance relationships of counts on upland and Pima cottons and on weekly and biweekly irrigated cotton were compared using analysis of covariance (SAS Institute 1988) to test for heterogeneity in regression parameters. We used Kuno's (1969) method to calculate fixed-precision sequential sampling stop lines from Iwao's mean–variance model:

$$T_n \geq (\alpha + 1)/(D^2 - (\beta - 1)/n) \qquad (1)$$

where T_n is the critical cumulative count over n samples, and D is precision, measured as SEM/m. Kuno's stop line is subject to the restriction that $n > (\beta - 1)/D^2$. Similarly, we used Green's (1970) method to calculate sampling stop lines from Taylor's mean/variance model as:

$$T_n \geq (an^{1-b}/D^2)^{1/(2-b)} \qquad (2)$$

Sample Plan Validation. We evaluated the performance of these sequential plan stop lines using counts from top-stratum leaves collected on two separate dates in 1992 (60 leaves per cultivar per date) and from counts on fifth mainstem node leaves on nine dates in 1993 (75–90 leaves per date). The samples in 1992 were those collected to study within-plant distribution patterns. Data from 1993 included counts from two of the dates on which within-plant distributions were examined plus counts from seven additional sample dates in the same 0.4-ha field. Mean densities in those samples ranged from 2.0 to 50.3 adults per leaf. We first examined the accuracy with which Taylor's power law and Iwao mean crowding regression predicted the sample variances of our 13 independent data sets. Then to assess the correspondence between actual and prescribed precision (SEM/mean), we used a resampling simulation technique modified from Hutchison et al. (1988) and Hutchison (1994). Basically, the simulation randomly selected successive samples without replacement from a given data set until the stop line criteria were met. Mean density (T_n/n), sample size, and precision were then calculated. The simulation was repeated 500 times for each data set, after which the distribution of precision values was formulated, and mean precision, mean density, and mean sample size values were calculated. Simulations were performed for each of the 13 data sets for each sequential sampling plan. The main limitation of this technique is that the validation data set must contain at least as many observations as needed in the most extreme cases (e.g., high desired precision, low mean densities). Because our validation data set was limited to 60–90 observations, we limited our validation to a desired precision value of 0.25 and densities ≥1 adult per leaf. Based on validation results, we then used simulation to refine the sampling plan for field implementation. In all analyses the minimum sample size was fixed at 10. A copy of the resampling simulation software is available upon request to S.E.N.

For comparative purposes we used the Monte Carlo approach of Nyrop & Binns (1991) to test Green's plan. The overall operation is very similar to the resampling simulation above, except that samples are not drawn from an actual data set but from a negative binomial distribution with a $k = m^2/[S^2\text{-}m^2]$ parameter calculated using S^2 from Taylor's power law for a given mean. We performed 500 iterations of the simulation for means ranging from one to 50 adults per leaf. Again, the minimum sample size was set at 10.

Insect Identity. Voucher specimens were submitted to A. C. Bartlett (USDA–ARS, Phoenix, AZ) both years of the study for identification. Based on a RAPD-PCR assay (Gawel & Bartlett 1993), the DNA pattern of these specimens was typical of that displayed by *B. tabaci* strain B.

Results and Discussion

Within-Plant Distributions. The density of adult *B. tabaci* differed significantly between strata ($F = 208.4$; df = 2, 472; $P < 0.01$) and was consistently highest on mainstem leaves from the top stratum of cotton plants in 1992 (Fig. 1A and B). The density of adults increased over time ($F = 711.3$; df = 1, 236; $P < 0.01$) and was greater on 'ST-506' than on 'DP-50' ($F = 57.8$; df = 1, 236; $P < 0.01$). The distribution of adults within the plant did not vary between cultivars ($P > 0.05$), but did vary over time ($F = 14.9$; df = 2, 472; P

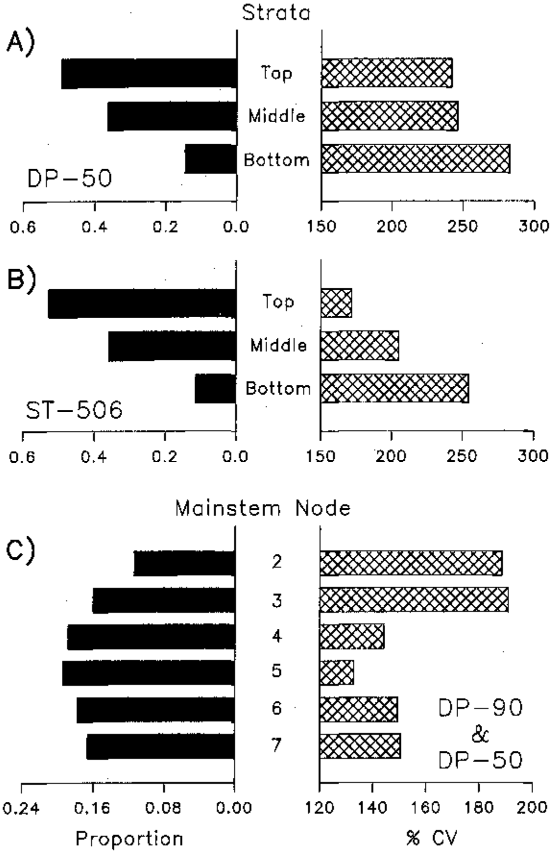

Fig. 1. Distribution of adult *B. tabaci* along the mainstem leaves of cotton plants and associated coefficients of variance (% CV = 100[SD/mean]) for three cotton cultivars. (A, B) Counts from the top, middle, and bottom strata leaves in 1992. (C) Counts on leaves from the first six nodes below the terminal in 1993. Distributions are based on total counts over four dates in 1992 and three dates in 1993.

< 0.01). The three way interaction between time, strata, and cultivar was not significant ($P > 0.05$). Changes in distribution between the different strata over time resulted from the change in the lowest density of adults from middle to lower strata leaves from the first to the second sampling date. The relative variation of counts pooled over all dates (% CV = 100[SD/mean]) was inversely related to abundance, with the lowest CVs being associated with the highest densities of adults on leaves near the top of the plant (Fig. 1A and B).

Similarly, in 1993 the density of adults increased over time ($F = 258.72$; df = 2, 241; $P < 0.01$) and differed among mainstem leaves ($F = 5.11$; df = 5, 1,205; $P < 0.01$ [Fig. 1C]). Further, a significant interaction between leaf and date indicated that the distribution of adults within the plant varied over time ($F = 10.58$; df = 10, 1,205; $P < 0.01$). The average nodal position of the most infested leaf was (mean ± SD) 4.9 ± 1.5, 5.5 ± 1.4, and 4.0 ± 1.4 at 1,262, 1,466, and 1,807 degree-days (12.8/30°C [Allen's (1976) method]) after planting, respectively. Adults were most abundant on leaves from the fifth mainstem node when observations were pooled over the three dates. The distribution, however, was relatively uniform, with roughly similar densities of adults on leaves from nodes 3 through 7. As in 1992, the lowest relative variation was associated with the highest density.

The within-plant distribution patterns that we observed are typical of those described for *B. tabaci* on cotton (Butter & Vir 1990, Rao et al. 1991, Naik & Lingappa 1992, Liu et al. 1993) and poinsettia (Liu et al. 1993). In general, adults are most abundant on younger leaves near the top of the plant and successively less abundant on leaves farther from the top of the plant. This pattern essentially mirrors the distribution of eggs over the whole plant (Butter & Vir 1990, Rao et al. 1991). On the top third of the plant, eggs are found in greatest abundance on the third mainstem leaf from the terminal (Naranjo & Flint 1994), whereas adults appear to be most abundant on the fifth mainstem leaf. However, these seemingly disparate distributions coincide when we consider the relatively flat distribution pattern of adults on this portion of the plant (Fig. 1C) and the decreasing size of leaves closer to the terminal. Thus, adults are actually more concentrated, on a per unit area basis, on leaves closer to the terminal.

Further analysis of within-plant distribution patterns was conducted by fitting Taylor's power law to counts from the three plant strata in 1992 (Fig. 2A). Analysis of covariance indicated no significant differences in the slope ($F = 0.21$; df = 2, 30; $P = 0.81$) or intercept ($F = 0.60$; df = 2, 30; $P = 0.56$) parameters of Taylor's power law regressions from the three strata. Thus, although densities differed significantly between leaves from the three strata, levels of aggregation did not differ. Fit of the power law to data from all three strata yielded (±SEM) $b = 1.53 \pm 0.04$ and $a = 2.12 \pm 1.09$ with $n = 39$. Likewise, results from 1993 also suggest that aggregation patterns of adults on leaves from the first six nodes below the mainstem terminal could be described by a single regression (Fig. 2B). Too few data points were available to fit separate power law regressions for each node; therefore this assertion could not be tested statistically. A fit of the power law to all nodes combined yielded (±SEM) $b = 1.78 \pm 0.06$ and $a = 1.42 \pm 1.11$ with $n = 18$.

In contrast to our findings, Tonhasca et al. (1994) found that adult *B. tabaci* displayed different degrees of aggregation between terminal and crown leaves of cantaloupe. This pattern could be related to the greater physical difference between terminal and crown leaves in comparison with that between cotton leaves of differing ages. As a result, sequential stop lines generated for different leaves of cantaloupe vary whereas a single stop line potentially could be used for any of a number of

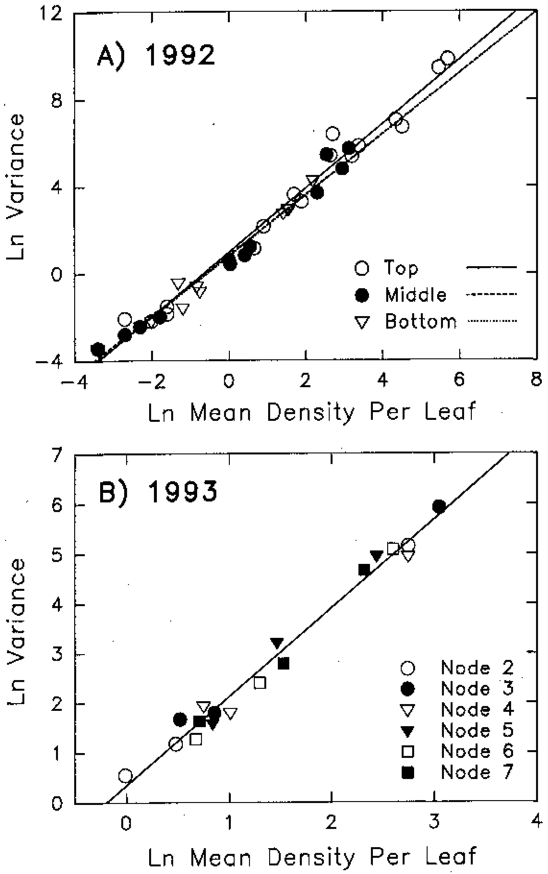

Fig. 2. (A) Taylor's power law regressions of mainstem leaf counts of adult *B. tabaci* from the top, middle, and bottom strata of cotton plants in 1992. (B) Relationship between *ln*(mean) and *ln*(variance) of mainstem leaf counts from the first six nodes below the terminal in 1993. Regression line is Taylor's power law fit to data from all nodes pooled.

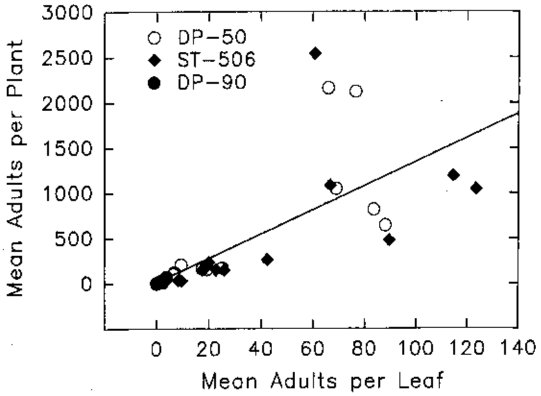

Fig. 3. Relationship between counts from leaves in the top stratum (1992: 'DP-50' and 'ST-506') or from fifth mainsten node leaves (1993: 'DP-90') and whole plant counts of adult *B. tabaci*.

leaves that might be selected as the sample unit on a cotton plant. The lower relative variation associated with counts from the fifth mainstem node leaf was the reason we selected that leaf as the sample unit. Thus, although that leaf had the highest density of adults on average, fewer samples would be necessary to achieve estimates with the same precision compared with counts from other leaves with lower densities of whiteflies. A similar rationale led to selection of that leaf for estimating the density of eggs and nymphs (Naranjo & Flint 1994).

Estimation of Absolute Density. There was considerable variation in the ratio between counts of adults on leaves from the top stratum of the plant or from fifth mainstem node leaves, and counts on whole plants. Over both years the number of adults per plant ranged from 0 to 2,541 and averaged (±SD) 282 ± 574. The mean ratio between top leaves and whole plants averaged 0.075 ± 0.071 over both years. Linear regressions relating leaf to whole plant counts did not vary significantly between DP-50 and ST-506 (slope: F = 2.42, df = 1, P = 0.13; intercept: F = 0.03, df = 1, P = 0.87). A simple linear regression of whole plant counts (y) on leaf counts (x) for all cultivars combined yielded $y = 13.34x + 15.20$ (F = 67.06; df = 1, 50; $r^2 = 0.58$; $P < 0.01$; SEM slope = 1.63; SEM intercept = 63.90) (Fig. 3). The intercept did not differ significantly from zero. The addition of degree-days from planting (12.8/30°C [Allen's (1976) method]) as an independent variable did not significantly (F = 2.23, P = 0.15) improve the predictive value of the regression.

Part of the variability in our results can be attributed to high plant-to-plant variation and the fact that we collected individual leaf samples and whole plant counts from different plants. A high degree of variability between leaf and whole plant counts was also reported by von Arx et al. (1984) for fourth instar *B. tabaci* on cotton. They chose to describe the ratio of leaf to whole plant counts as a curvilinear function of accumulated degree-days. This physiological measure of time did not improve predictions of absolute density in our study. Aside from the regression constant, using either the ratio or the linear regression model produced almost the same estimates of absolute density.

Within-Field Distributions. Of 64 data points collected at the MAC in 1993, 59 had means >0 and were usable for calculating parameters for Taylor's power law and Iwao's mean crowding regression (Table 1). Analysis of covariance indicated that neither the slope nor intercept parameters were significantly different between cultivars or between irrigation regimes for either mean–variance model ($P > 0.10$). Thus, we estimated parameters for both models using a pooled data set (Table 1). Fits of both models yielded high values of r^2. Both b and β were significantly >1 indicating contagious

Table 1. Regression statistics and parameter estimates for Taylor's power law and Iwao's mean crowding regression, Maricopa, AZ, 1993

Treatment	n	Power law			Mean crowding regression			Density range
		a (SE)	b (SE)	r^2	α (SE)	β (SE)	r^2	
'DP-50'	28	2.05 (1.14)	1.66 (0.06)	0.97	−0.42 (0.69)	1.98 (0.06)	0.97	0.1–29.5
'Pima S-7'	31	2.11 (1.16)	1.69 (0.07)	0.96	−0.74 (0.75)	2.11 (0.07)	0.97	0.1–21.4
Weekly irrigation	30	1.95 (1.16)	1.66 (0.08)	0.95	−0.62 (0.77)	2.04 (0.09)	0.95	0.1–17.4
Biweekly irrigation	29	2.26 (1.14)	1.68 (0.05)	0.98	−0.42 (0.94)	2.02 (0.07)	0.97	0.1–29.5
Combined	59	2.08 (1.10)	1.67 (0.05)	0.97	−0.53 (0.58)	2.03 (0.05)	0.96	0.1–29.5

or aggregated distributions for adults using the fifth mainstem node leaf as the sample unit.

Sequential Sample Plan. Following Green's (1970) and Kuno's (1969) methods, we calculated sequential sampling stop lines for adult *B. tabaci* at two levels of precision (SEM/mean) (Fig. 4). Prescribed sample sizes differed somewhat between the two methods. For instance, at $D = 0.25$ Green's plan would require 35 samples at a mean density near one, but Kuno's plan would require only 25 samples. At the same density but with $D = 0.1$ Green's plan would require 200 samples, 40 more than Kuno's plan. Both plans prescribed roughly similar sample sizes at densities between one and 10; however, because of the restriction that $n > (\beta\text{-}1)/D^2$ in Kuno's plan, a minimum of 17 samples is required at densities >15 with $D = 0.25$, and a minimum of 103 samples would be required at densities >45 with $D = 0.1$. This limitation could be important in field application of Kuno's plan. The selection of the best method should rest on the accuracy of each sampling model and the performance of each plan against independent field data.

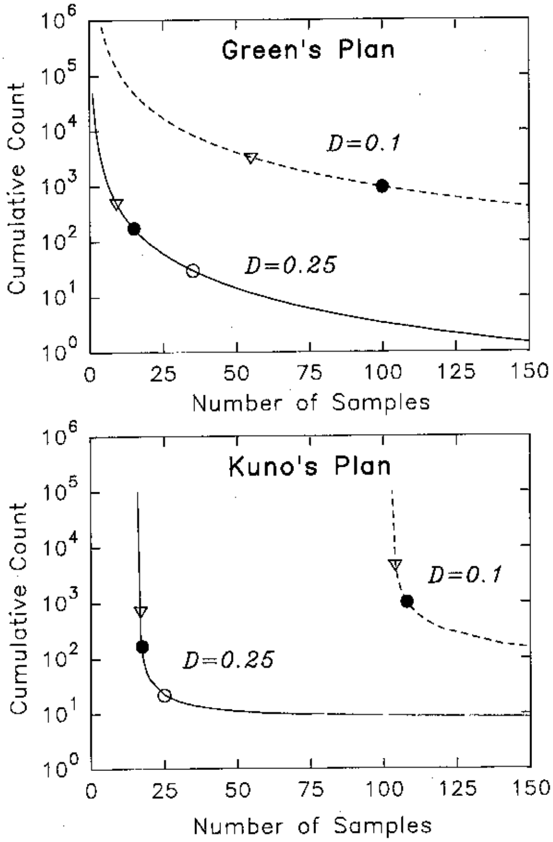

Fig. 4. Sequential sampling stop lines for estimating the density of adult *B. tabaci* at fixed precisions (SEM/mean) of 0.1 and 0.25 based on Green's (1970) and Kuno's (1969) methods. The sample unit is a fifth mainstem node leaf counted from the terminal (=node 1). Open circles, solid circles, and open triangles denote mean densities of 1, 10, and 50 adults per leaf, respectively.

Sample Plan Validation. We first examined the accuracy of each sampling model in predicting variances observed in our independent data sets (Fig. 5). Our independent field data sets covered a range of densities from 2.0 to 50 adults per leaf and a period of time from 16 July to 15 October (Tables 2 and 3). These densities and time periods are representative of those over which the sample plan would be utilized in central Arizona. For both models, slopes of the regressions between predicted and actual variance did not differ significantly from one ($P > 0.05$). Taylor's power law overestimated and underestimated the variance in roughly equal proportions whereas the mean crowding model was biased and consistently overestimated the variance.

The performance of fixed-precision sequential sample plans depends on the accuracy of the underlying model that predicts the sample variance from the sample mean. Many factors are known to affect these model parameters, including host crop, geographic range, and environmental heterogeneity (e.g., Trumble et al. 1989, Jones 1990). We found that neither Taylor's power law nor Iwao's mean crowding regression accurately predicted the variance of independent samples taken from the same site in different years or from sites in relatively close proximity to one another in the same

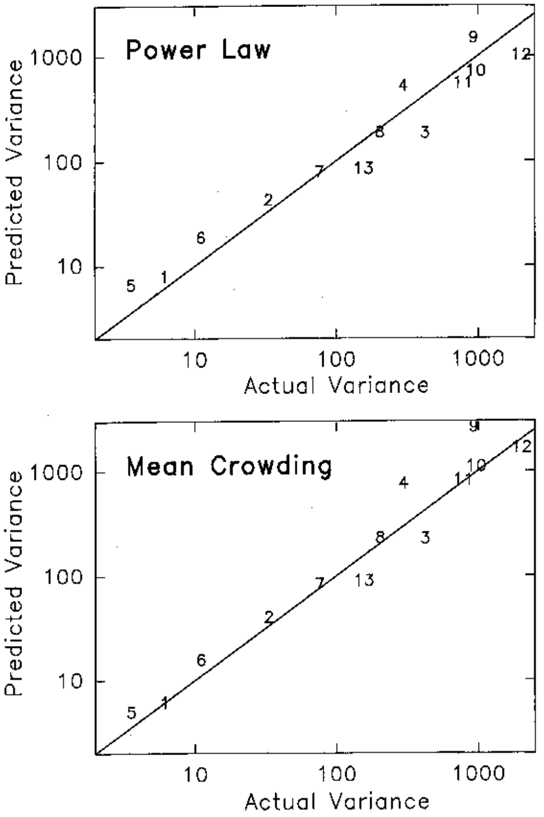

Fig. 5. Prediction of variances from independent validation data sets using Taylor's power law and Iwao's mean crowding regression to model the mean–variance relationship. Numbers refer to data points listed in Tables 2 and 3. The lines represent $x = y$.

year. This reflects the fact that the parameters of mean variance models are not fixed, species-specific descriptors, but are instead samples from a population of parameter values for the species (e.g., Jones 1990). However, from the perspective of sample plan implementation, the important question is whether our selected parameters will yield estimates of population density with acceptable precision.

The desired precision specified in a sample plan is not the precision that one will achieve from any one sample, but instead is the expected precision over a large number of sampling bouts (Hutchison et al. 1988, Nyrop & Binns 1991). Several approaches are available for assessing the expected performance of sequential sampling plans in the field. Monte Carlo simulation (Nyrop & Binns 1991) can be used; however this approach requires the assumption of a specified distribution. A more robust procedure is to use actual data sets collected independently from those used to develop the sample plan (Hutchison 1994). The greatest strength of this approach is that the underlying distribution is defined by the data set rather than by a theoretical model. By resampling these data sets on a computer, we can define the average behavior of the plan for that data set given that the individual observations from a field could have been collected in any order. We examined the performance of our sample plan using both methods.

Based on our resampling analyses, sample size requirements were similar for Green's and Kuno's methods at densities below ≈15 adults per leaf (Tables 2 and 3). Above that density, Green's plan required fewer samples for roughly the same levels of precision. This resulted because of the minimum sample size limitation for Kuno's plan discussed above. Neither plan consistently achieved the prescribed precision of 0.25. Thus, based on the 1992 data sets each plan required more samples than necessary to estimate population density with the desired precision. Results from the 1993 data sets were more variable. Green's plan equaled or exceeded the desired precision on the first five dates and failed to meet the desired precision on the final four dates. Kuno's plan exceeded the desired precision on the first seven dates and failed to achieve the desired precision on the final two dates. These results are a direct consequence of the accuracy with which Taylor's power law and the mean crowding regression predicted the variances of these samples (Fig. 5). Finally, the distribution pattern of actual precision values varied considerably for both plans. However, despite the failure to achieve prescribed precision in two and five instances for Kuno's and Green's plan, respectively, in only three of those cases did actual precision values exceed 0.325 more than 40% of the time (Tables 2 and 3). A simple average of mean precision values over all 13 data sets yielded 0.24 for Green's plan and 0.22 for Kuno's plan.

In contrast with our resampling method, Monte Carlo validation (Nyrop & Binns 1991), which assumed a negative binomial count distribution, suggested that Green's plan performed very well. At densities ranging from 1 to 50 adults per leaf, Green's plan achieved average precisions that steadily declined from 0.248 to 0.221. Further, actual precision was poorer than 0.325 in only 11 to 16% of the simulations at any density. The better than prescribed precision at higher densities was largely a function of the fixed minimum sample size of 10 leaves.

Contrasting results from using both approaches in our analysis suggest that distributional patterns of adult *B. tabaci* may not be adequately defined by a negative binomial model, at least over the range of densities examined. Similar results were found by Hutchison (1994) in comparing the two approaches for validating sampling plans for *Acyrthosiphon pisum* (Harris) on alfalfa. Monte Carlo simulation can be a useful tool for sample plan validation if careful attention is given to the selection of an appropriate distribution model. Still, the evaluation of the sample plan in the field is clearly

Table 2. Performance of Green's sequential sampling stop lines on independent data sets collected in 1992 and 1993

Data set	Data set		Simulation			Distribution of precision				
	Estimated density	n	Mean density	Mean sample size[a]	Mean precision	<0.175	0.175–0.225	0.225–0.275	0.275–0.325	>0.325
1992 ('DP-50')										
(1) 16 July	2.2	60	2.3	26	0.21	0.13	0.46	0.37	0.03	0.00
(2) 29 July	14.5	60	15.0	15	0.27	0.34	0.20	0.03	0.09	0.35
1992 ('ST-506')										
(3) 16 July	6.0	60	6.2	19	0.22	0.08	0.55	0.36	0.02	0.00
(4) 29 July	27.0	60	27.5	12	0.18	0.41	0.50	0.09	0.00	0.00
1993 ('DP-90')										
(5) 30 July	2.0	90	2.0	27	0.18	0.31	0.68	0.01	0.00	0.00
(6) 12 Aug.	3.7	90	3.7	22	0.19	0.35	0.54	0.10	0.002	0.00
(7) 17 Aug.	8.7	75	8.9	17	0.23	0.09	0.42	0.33	0.12	0.03
(8) 30 Aug.	14.7	75	15.1	14	0.25	0.02	0.25	0.44	0.27	0.03
(9) 7 Sept.	50.3	75	49.9	10	0.18	0.43	0.41	0.15	0.01	0.00
(10) 17 Sept.	32.5	75	33.5	11	0.28	0.03	0.15	0.32	0.31	0.18
(11) 22 Sept.	27.9	75	27.6	12	0.28	0.02	0.16	0.27	0.32	0.24
(12) 1 Oct.	40.0	75	40.0	11	0.33	0.00	0.05	0.17	0.27	0.51
(13) 15 Oct.	9.1	75	9.5	17	0.33	0.00	0.01	0.08	0.35	0.57

Summary statistics are based on 500 iterations of the resampling simulation with precision $D = 0.25$. Numbers preceding sample dates refer to points graphed in Fig. 5.

[a] Minimum sample size of 10 specified in all simulations.

the best test of the overall performance and value of the plan.

Sample Plan Selection and Refinement. Because Green's plan performed better, on average, in terms of achieving the desired precision and because the plan does not force an unreasonable minimum sample size, particularly at higher densities, we selected this plan for further refinement. Additional tests were conducted with our resampling simulation to more closely match actual with prescribed precision. With some exceptions, Green's plan required more samples than necessary at relatively low densities and too few samples at relatively high densities. There were several clear exceptions to these trends (e.g., 15 October and number 4, 29 July); however, our data set was not extensive enough to separate the positively correlated aspects of density and time of season. Thus, we made preliminary adjustments according to two density classes: <15 and >15 adults per leaf. On

Table 3. Performance of Kuno's sequential sampling stop lines on independent data sets collected in 1992 and 1993

Data set	Data set		Simulation			Distribution of precision				
	Estimated density	n	Mean density	Mean sample size[a]	Mean precision	<0.175	0.175–0.225	0.225–0.275	0.275–0.325	>0.325
1992 ('DP-50')										
(1) 16 July	2.2	60	2.2	21	0.21	0.06	0.32	0.40	0.20	0.02
(2) 29 July	14.5	60	15.0	18	0.26	0.37	0.12	0.06	0.12	0.33
1992 ('ST-506')										
(3) 16 July	6.0	60	6.1	18	0.22	0.09	0.45	0.39	0.06	0.002
(4) 29 July	27.0	60	26.9	17	0.15	0.84	0.16	0.002	0.00	0.00
1993 ('DP-90')										
(5) 30 July	2.0	90	2.0	21	0.21	0.11	0.59	0.29	0.01	0.00
(6) 12 Aug.	3.7	90	3.7	19	0.20	0.23	0.48	0.27	0.02	0.00
(7) 17 Aug.	8.7	75	8.9	18	0.22	0.12	0.46	0.29	0.11	0.02
(8) 30 Aug.	14.7	75	14.8	17	0.23	0.05	0.40	0.44	0.10	0.01
(9) 7 Sept.	50.3	75	50.3	17	0.15	0.89	0.11	0.00	0.00	0.00
(10) 17 Sept.	32.5	75	32.4	17	0.22	0.09	0.44	0.40	0.06	0.01
(11) 22 Sept.	27.9	75	28.0	17	0.24	0.04	0.37	0.41	0.17	0.01
(12) 1 Oct.	40.0	75	39.6	17	0.27	0.01	0.17	0.40	0.32	0.11
(13) 15 Oct.	9.1	75	9.3	18	0.32	0.00	0.01	0.18	0.37	0.44

Summary statistics are based on 500 iterations of the resampling simulation with precision $D = 0.25$. Numbers preceding sample dates refer to points graphed in Fig. 5.

[a] Minimum sample size specified as the greater of 10 or the solution to $n > (\beta - 1)/D^2$ (see Kuno 1969) in all simulations.

average, prescribed precisions of 0.30 and 0.20 generated stop lines (equation 2) that resulted in actual precisions near 0.25 when densities were <15 and >15 adults per leaf, respectively. A more extensive validation database is needed to more accurately refine our sampling plan.

At currently proposed action thresholds of 5–10 adults per leaf for *B. tabaci* in Arizona (Ellsworth & Meade 1994), our plan would require <20 leaf samples to estimate these densities with a precision of 0.25. We are currently focusing our efforts on improving sampling efficiency even further by developing plans based on binomial count models (Naranjo et al. 1994) for classifying whitefly populations for pest management decision-making. Our fixed-precision sequential sampling plan allows for the efficient estimation of adult *B. tabaci* population density in cotton and should prove useful for research and management purposes.

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