Spatial Distribution and Sampling of *Aulacaspis yabunikkei* (Homoptera: Diaspididae) in Camphor Trees

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ABSTRACT

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Dispersion patterns of varied stages generated by Iwao's patchiness regression and Taylor's power law for *Aulacaspis yabunikkei* Kuwana in different sampling units of camphor trees, *Cinnamomum camphora* (L.), were compared. Taylor's power law provided a consistently good fit to the data, whereas the fit of Iwao's patchiness regression were erratic, and the values of aggregation index of Taylor's power law (1.76 to 2.65) were narrower than those of Iwao's (1.26 to 11.83), but both indices (b > 1.7 and β > 1.2) indicate a clumped distribution pattern in all sampling units. Mean numbers of scales per leaf differed significantly (P < 0.05) between the lower and upper layer of the canopy, between old and young leaves, and between the underside and upper surface of leaves. The number of eggs corresponded the most closely to the total population, and this stage is best for precisely estimating the population. However, for the non-professional, the eggs are difficult to count. The number of female adult scale covers were easy to count and also corresponded very closely to the total population. Thus, female adult scale covers on the under leaf surfaces of old leaves were chosen as sampling targets to represent the scale population and for the decision making of pest management. Scales only with high density would damage camphor tree heavily, therefore, in fixed sampling plan, when there were 100 female

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scales on underside of leaf per twig in a tree, the optimum samples of 12 twigs per tree and 3 twigs per tree would reach 0.25 and 0.5 precision levels, respectively.

(Key words: *Aulacaspis yabunikkei*, spatial distribution, sampling)

INTRODUCTION

White cinnamomum scales, *Aulacaspis yabunikkei* Kuwana, has been a serious pest of camphor trees, *Cinnamomum camphora* (L.), for decades, and camphor tree is the most popular ornamental avenue tree in Taiwan. This insect could make camphor trees death when damaged heavily, therefore it is an important pest. Despite its importance, yet only a few studies[12, 14, 15, 16] has been done before 1970, and after 20 years, only Lee and Peng[7] (1992) studied this insect's damage and control. Thus basic information of its ecology remains insufficient. A good control of pest is based on its basic ecology information, therefore we need more studies on this insect.

Management of diaspidid scales relies on the use of insecticides or pruning cuts. The spatial distribution of scales determine the appropriate cutting area and also give information for making sound decisions on insecticide treatment. Also, knowing the distribution of insect counts can contribute to a sound sampling program[11, 13]. This program can be used to estimate or classify precisely the population density of scales.

Excepting crawler stage, the male adult was the only moving stage, so for sessile female with larger size and for laying more eggs, it should need more resource and it should be more dispersal than male. Koteja[6] pointed that male and female crawlers sometimes have different wandering and settling behaviours and male crawlers exhibit a gregarious instinct in some species. The different spatial distribution between sex in white cinnamomum scales would be calculated. Though investigating the crawler stage distribution could directly understand the different spatial distribution between sex, the differences are observable only in slide-mounted specimens. From the second stage, the sexual dimorphism observed available. To explore whether scales of different sexes have different dispersal patterns, the spatial distribution of males and females of various stages from second stage of *A. yabunikkei* on camphor trees were compared. Meanwhile, a sampling plan was proposed to provide better control for *A. yabunikkei*, so to determine the best sampling unit and representative stage are necessary to develop optimal sample sampling plan.

MATERIALS AND METHODS

The insects

To investigate population densities and hence to explore the population and its impact factors of white cinnamomum scales, an experimental plot of 260 trees along one side of a 2.4-km stretch of Tz-nan Rd. in Taipei City was sampled weekly from April 1994 to March 1995. Tz-nan Rd. is a mountain street, therefore, only one side can be planted with camphor tree. These trees were in the same age, and were not influenced by pesticide spraying or prunning during the sampling period. Twenty trees were labeled randomly in each sampling day and each tree's circumference of trunk at 150 cm above the ground was also measured. The canopy of each tree was divided into 2 parts, the sunshine-receiving upper layer and the shaded lower layer; three twigs with leaves about 30 cm in length were picked each from upper and lower parts, placed into plastic bags, and brought back to the laboratory.

Samples were preserved at 4 ℃ and examined within a week. The scale coverings of all sampled individuals were removed and the scales were examined using a Wild M8 binocular microscope. The sampled insects were categorized by the following procedure: if the bodies were complete but the color had changed from yellow to black or they were dry, then they were designated as death; the presence of fluid after the bodies were punctured signified a live scale insect; scales being incomplete or with parasitoid emerging pores or parasitoids under the scale were designated unnatural death. Numbers of 2nd or 3rd female instars were counted, and numbers of individuals of living, natural death, or unnatural death of each stage were recorded, respectively. The number of eggs and crawlers were directly counted, the male nymphs (2nd, 3rd, and 4th instars) were counted by scales or the scales removed when hardly discriminated. Complete male scales of whitish coloration were treated as live individuals, and those damaged scales were treated as dead ones.

Spatial distribution

The scale numbers are calculated by per leaf, per twig and per tree, respectively. We calculated the mean and variance of scale numbers on leaves per twig to estimate the spatial distribution among leaves, on twigs per tree to estimate the spatial distribution amnog twig, and on trees per investigative time to estimate the spatial distribution among trees. Differences between two commonly used dispersion indices, Taylor's power law[17] and Iwao's regression[5] are calculated and compared. Those were applied to the mean number of insects among trees, twigs, or leaves to estimate the spatial distribution of white cinnamomum scales in camphor trees.

Taylor's power law is as follows: $s^2 = am^b$; where s^2 is the variance of the number of scales in sampling units, and m is the mean number of scales in sampling units. The coefficients a and b were estimated from the following regression equation[13]:

$$Log\ s^2 = Log\ a + b\ (Log\ m) \qquad (1)$$

Where a is the sampling factor that changes with the sampling unit, and b is the dispersion index of a certain species.

The regression method of Iwao's, i.e., Iwao's patchiness regression, was calculated by solving the equation:

$$\overset{*}{m} = \alpha + \beta m \qquad (2)$$

where α is a basic individual unit for spatial distribution, and β is the clumping index of individual spatial dispersion. Mean crowding, $\overset{*}{m}$, was derived from Lloyd's formula[8]:

$$m^{*} = m+(s^2/m)-1 \quad (3)$$

and by substituting the mean and variance from the counted data.

Development of the sampling plan

Appropriate sampling unit, i.e., young or old leaves, the underside or upper surface of the leaves and representative stage were determined for deveolping sampling plan of white cinnamonum scales in camphor tree.

(1) *Sampling unit.* Three twigs with leaves about 30 cm length were randomly sampled separately from the sunshine-receiving upper layer and the shaded lower layer of the trees as above description. Then, insect scales of each stage (including scale residues) on the underside or upper surface of old, dark green leaves or young, light green leaves were counted.

The data collected to study the among-twig distribution that was originally separated by leaf-age was used to compare the 2 population measures, i.e., the number of scales per young leaf and the number of scales per old leaf. Furthermore, the data collected to study the among-leaf distribution was used to compare the 2 population measures, i.e., the number of scales of underside of leaf per twig and the number of scales of upperside of leaf per twig. The curves of the 2 populations were drawed by linear regression over population per leaf and compared. Appropriate sampling unit was then determined based on its correlation to the whole population.

(2) *Representative stage.* Linear regressions of all living individuals per twig on living individuals of eggs, crawlers, 2nd nymphal females, female adults, male nymphs, and female adult's scales (including scale residues) on the determined unit per twig were calculated, respectively. After linear regression analysis, based on the simplicity of discrimination and counting and its representative to scale population, an appropriate representative stage was determined.

(3) *The fixed-sample sampling.* Wilson and Room (1982)[19] incorporated *a* and *b* indices derived from Taylor's power law to calculate the optimum number of intratree samples (N) necessary to estimate mean density (m) with a given level of precision (D) expressed as a proportion of the mean as follows:

$$\mathrm{N} = \left(Z_{\alpha/2} / D\right)^2 \cdot (S / m)^2 \quad (4)$$

Where Z is the standard normal deviate (α = 0.1 then it is 1.64 in two tails). Precision levels of D = 0.25 and 0.5, were chosen for the present study.

RESULTS

Spatial distribution

Dispersion indices in different stages are shown in Table 1. Taylor's aggregation indices of various stages among trees ranged from 1.78 to 2.65, and female nymph was the most aggregative stage while male nymph was the least. The indices of various stages among twigs ranged from 1.76 to 2.14, and egg was the most aggregative stage while female nymph was the least. The indices among leaves ranged from 1.62 to 2.47, and crawler was the most aggregative stage while female adult was the least.

Female nymph and female adult aggregated more with larger sampling areas but male nymph showed the opposite pattern.

Iwao's patchiness regression indices of various stages among trees ranged from 7.11 to 11.83, and egg was the most aggregative stage while female nymph was the least. The indices of various stages among twigs ranged from 1.55 to 2.05, and female adult was the most aggregative stage while crawler was the least. The indices among leaves ranged from 1.29 to 2.77, and male nymph was the most aggregative stage while female adult was the least. Only female adult aggregated more with larger sampling areas.

Taylor's aggregation indices have a narrower range with from 1.62 to 2.65 than Iwao's do with from 1.29 to 11.83. Taylor's power law consistently provided a good fit to the data (R^2 from 0.89 to 0.99), and Iwao's patchiness regression provided erratic fit to the data (with R^2 from 0.54 to 0.90). The relationships of Taylor's and Iwao's indices for variable-separated stages and sampling units were different, but all indices agree that each stage of *A. yabunikkei* is aggregated. It is also found that the aggregated indices of male nymphs are larger than those of female nymphs except among trees' distribution.

Table 1. Taylor's power law and Iwao's patchiness regression indices[1)] for *Aulacaspis yabunikkei* on trees, twigs, and leaves

Stage	Parameters of spatial distribution								
	Among trees (n = 50)			Among twigs (n = 978)			Among leaves (n = 2932)		
Taylor's power law									
	a	*b*	R^2	*a*	*b*	R^2	*a*	*b*	R^2
Egg	2.70	2.24	0.97	1.04	2.14	0.99	1.04	2.42	0.97
Crawler	2.08	2.31	0.97	1.02	2.12	0.98	1.02	2.47	0.97
Male nymph	13.52	1.78	0.95	1.02	2.13	0.99	1.03	2.46	0.97
Female nymph	1.23	2.65	0.94	0.96	1.76	0.93	0.99	1.76	0.92
Female adult	2.92	2.21	0.89	0.89	1.78	0.92	0.97	1.62	0.91
Total counts	3.67	2.06	0.95	0.91	2.07	0.97	0.99	2.29	0.96
Iwao's patchiness regression index									
	α	β	R^2	α	β	R^2	α	β	R^2
Egg	-88.56	11.83	0.70	14.48	1.56	0.82	5.09	1.74	0.63
Crawler	-10.62	8.30	0.90	5.70	1.55	0.87	1.53	2.75	0.76
Male nymph	-86.76	11.08	0.81	7.18	2.04	0.85	3.60	2.77	0.54
Female nymph	2.63	7.11	0.88	0.14	1.72	0.83	-0.02	2.49	0.83
Female adult	-34.46	10.02	0.68	-0.97	2.05	0.86	0.26	1.29	0.87
Total counts	-151.10	8.71	0.76	24.40	1.59	0.84	9.49	1.72	0.63

[1)]Taylor's power law and Iwao regression were all significant at the $P < 0.001$ level.

Development of the sampling plan

(1)*Sampling unit.* In our investigation, 46 trees and a total of 1538 leaves (13 ± 4 leaves per twig) in first two sampling were taken. Of the insects sampled, only 5% were in the upper canopy while 95% occupied the lower canopy. Thus, the population density on the lower canopy was significantly greater than that on the upper canopy (t = 1.69; df = 45; P < 0.05). In the following investigation period, sampling was confined to the lower canopy of each tree.

During the investigation, the trees's circumferences were about 21.0 ± 9.5 cm at breast height (N = 976). And there were 8.4 ± 0.3 leaves per twig on the average (N = 2929). There were 8.0 ± 2.9 old leaves and 0.5 ± 1.4 young leaves per twig on the average. There were significantly more insects on old leaves than on young leaves (t = 8.32; df = 3253; P < 0.01). Each young leaf had only 0.1 insects but there were 6 insects on each old leaf (Fig. 1). Thus old leaves were suggested to be the target of sampling.

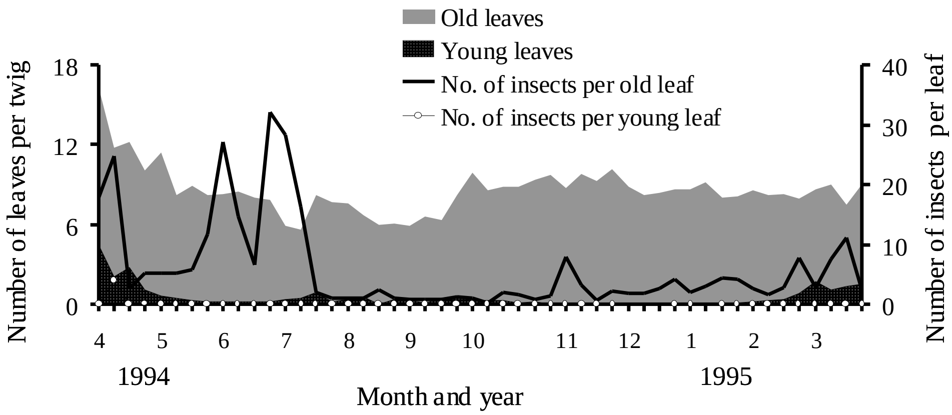

Fig. 1. Population densities of *Aulacaspis yabunikkei* on young or old leaves in camphor trees along Tz-nan Rd. in Taipei.

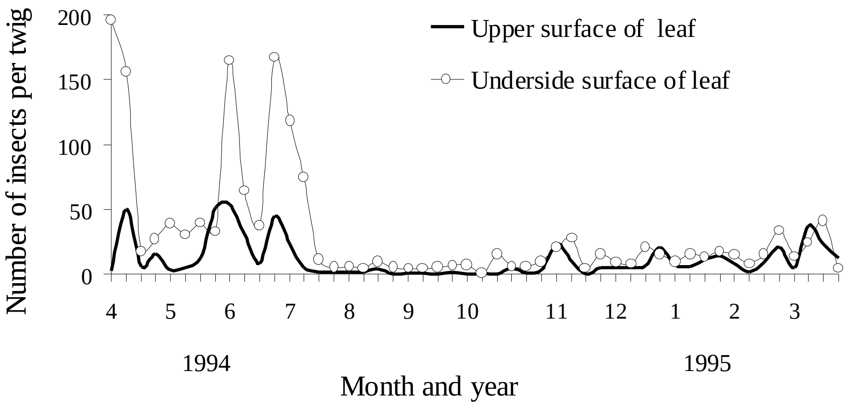

Fig. 2. The population fluctuation of *Aulacaspis yabunikkei* on upper, or under side surfaces of leaves of camphor trees.

A. yabunikkei distributed itself more on underside surfaces of leaves than on upper surfaces. The lower leaf surfaces had 74% of the scales (t = 4.92; df = 2928; P < 0.05). There were about 3 times the number of insects on underside surfaces of leaves than on upper surfaces from April to September, and densities were similar from November to March (Fig. 2). Insect scales on underside leaf surfaces (R^2 = 0.96) correlated with those on both leaf surfaces more closely than did those on upper surfaces (R^2 = 0.58). Therefore, underside leaf surfaces can be an appropriate sampling unit for *A. yabunikkei*.

(2)*Representative stage*. The correlation between eggs and total living insects (R^2 = 0.89) was higher than that for the other stages, and crawler was the second highest one (R^2 = 0.59) (Fig. 3).

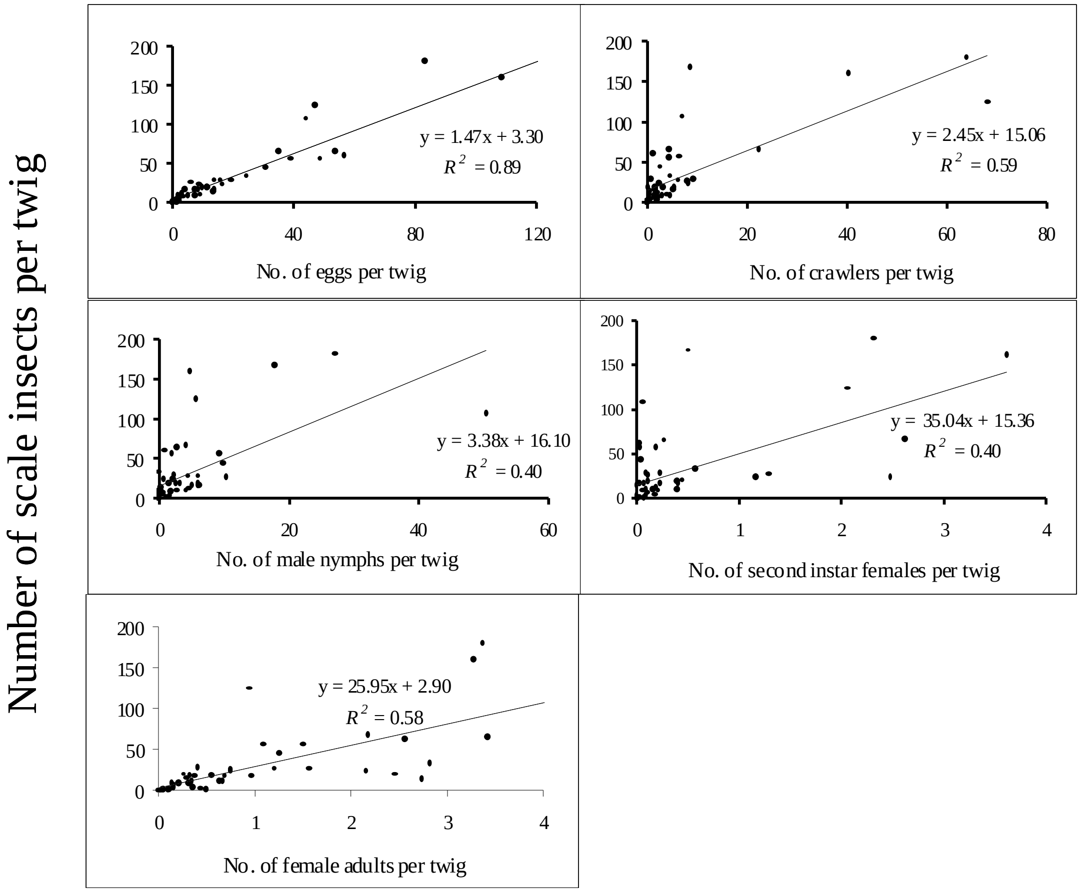

Fig. 3. Linear regressions of the sample mean densities of various stages to the total numbers of living *Aulacaspis yabunikkei* on camphor trees.

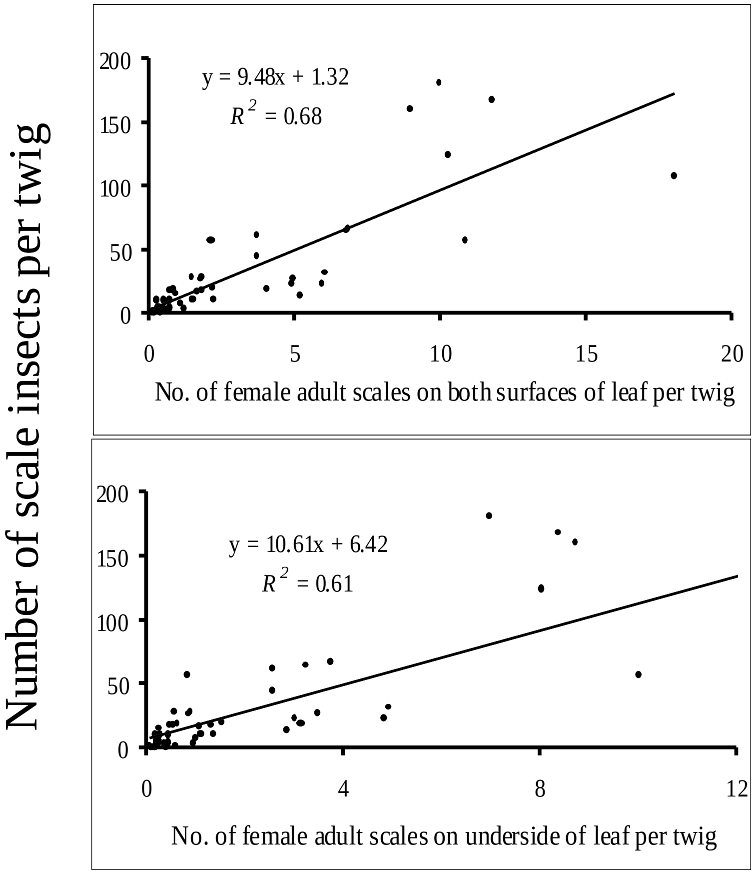

Fig. 4. Linear regressions of population densities of adult female covering scales on both leaf surfaces or just the under side surface of leaves to total densities of *Aulacaspis yabunikkei*.

Directly counting the covering scales of adult females without determining if she was alive was much easier, and there is a significant correlation between covering scales of adult females and all living insects on the target-sampling units (Fig. 4). The correlation between covering scales of adult females on both leaf surfaces or underside surface and all living insects on the target-sampling units both were higher than that of the crawler ($R^2 > 0.60$). Therefore, the scale numbers of female adults on the underside old leaves per twig were suggested as the sampling target.

(3)*The fixed-sample sampling*. Since Taylor's power law regression fitted the data better than Iwao's patchiness regression. The optimal sample size has thus been determined on the assumption of Taylor's power law. The *a* and *b* indices of above target sampling were 0.92, and 1.74, respectively, and there were 0 to 636 female adults on the underside old leaves per twig in our investigation. According to Equation (4), the optimum samples of 33 twigs per tree with mean number above 2 scales per twig (α = 0.1) and 8 twigs per tree for the same condition would reach 0.25 and 0.5 precision levels, respectively (Fig. 5). During the investigated period, it was found that 296 scales per twig in a tree would make the tree die in the field. If the 1/3 of above density was assumed as damage density, the optimum samples of 12 twigs per tree with mean numer above 100 scales per twig and 3 twigs per tree for the same condition would reach 0.25 and 0.5 precision levels, respectively .

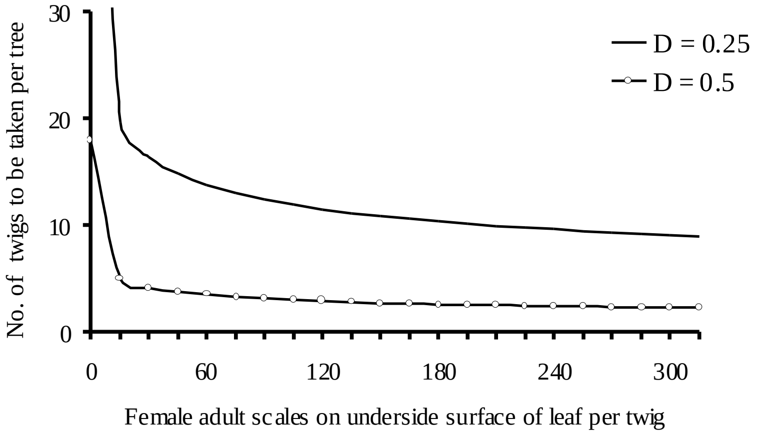

Fig. 5. Optimal number of samples to be taken to estimate mean population densities of *Aulacaspis yabunikkei* per twig using enumerative sampling procedures with reliability level of D = 0.25 and D = 0.5.

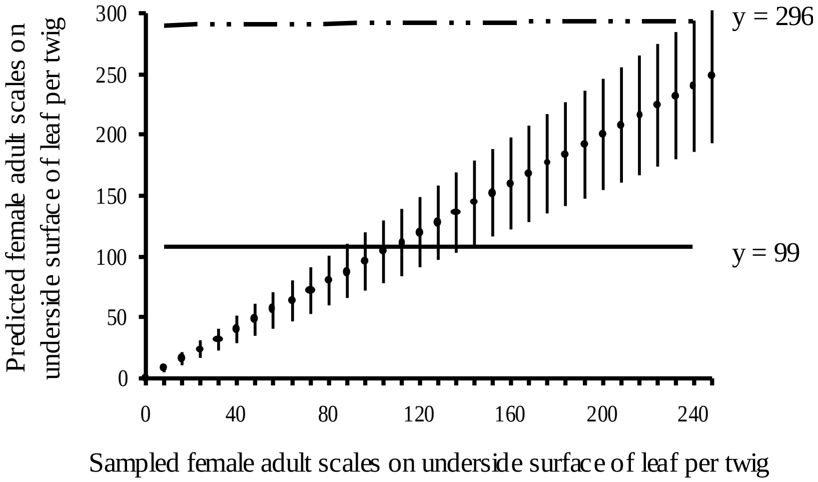

Fig. 6. The estimated mean densities with 0.5 precision levels of *Aulacaspis yabunikkei* per twigs with three twigs per tree.

In fixed-sample sampling, when we sampled 3 twigs per tree on average to estimate population density in the field, as scale's density increased from 1 to 300 scales per twig, the precision level were decreasing from 0.91 to 0.43 (Fig. 6). Thus, in low scale's density, the estimated density reached poor precision, while as density increased, the estimated density would reach higher precision level.

DISCUSSION

Spatial distribution

Eggs are only found under the mother's scale. As a result, the egg aggregation indices (*b*) are larger than 2 in all sampling units and the egg stage is the most aggregative in twig sampling unit. The crawler stage is mobile and can choose the settlement location, thus determines the aggregative distribution patterns of the subsequent stages. Though crawlers could leave the cover of their parent female, some behaviors of crawlers influence their distribution, e.g., crawlers display high thigmotactic behavior, negative phototaxis, and they also tend to settle closely to the parent female[4]. So the distribution of crawlers is still as aggregative as egg stage in all sampling units. The tendency of crawlers to aggregate (Table 1) is partly explained by the short distance crawlers tend to move and by their thigmotactic behavior [10]. During settlement, female crawlers may move 4 times the distance that male crawlers move from their mother scale, and males tend to settle closely to the parent female [4]. This explains why the aggregated indices, *b*, of male nymphs are larger than those of female nymphs (Table 1).

Our calculation of Taylor's power law and Iwao's regression coefficients for trees, twigs, or leaves supports the conclusion of recent simulation analyses that the magnitude of Taylor's *b* and Iwao's β appears to be sampling unit dependent[9]. Thus, it would be important to maintain a consistent sampling unit for comparative studies of distributional patterns[9]. Taylor's aggregation indices of this insect have a narrower range than Iwao's do and all of Taylor's linear regressions fit better than those of Iwao's. These results were predictable because the patchiness regression is not subject to the stabilizing effect of a log transformation[18].

Determine of the sampling plan

(1)*Sampling unit.* Like *Saissetia oleae* [3], *A. yabunikke* is also more common on the lower part of the canopy. Differences of microenvironments, e.g., different light characteristics and/ or temperatures may be key factors causing different densities between the upper and lower canopies. But it did need more researches to prove it.

In spring, the camphor trees grew more new leaves, so the mean numbers of new leaves were 1.2 ± 2.3 leaves per twig, while there were 0.2 ± 0.9 new leaves in other seasons. The time of old leaves remained on tree are longer than young leaves. Thus, the variance of young leaves' number was larger than that of old leave's. Though there were more insects on old leaves in our result, this test was not designed to prove preference between old and young leaves but to determine the most efficient sampling unit.

The proportion of insects on leaf surfaces changed with seasons. Crawlers' choice of leaf surfaces depended on adequate temperatures [1]. They would set more on upper surfaces of leaves in winter but less in summer. Based on the coefficient of determination (R^2) of the 2 regression lines, *A. yabunikkei* on underside surfaces of old leaves in the lower canopy of trees was selected as our sampling target.

(2)*Representative stage*. The correlation between eggs and total living insects was higher than that for the other stages, but eggs were too small to be counted with the naked eyes, and it is inconvenient to count them under a stereomicroscope. Using egg stage is only for the professional to estimate population precisely. When there were many covering scales of adult females on the both surfaces of leaves in the lower canopy, there were high total densities of *A. yabunikkei*. The best timing to control crawlers of the next generation can be estimated by counting the covering scales of female adults on the both leaf surfaces of old leaves. Since this procedure had a high representation and is also simple for manipulation, selecting this stage as the sampling target can presumably reduce counting errors and sampling costs, and allow non-professional personnel to conduct it.

(3)*The fixed-sample sampling*. The scales are much clump distribution, so the sample's strandard error is much larger than mean in our investigation. When the density reaching 296 scales per twig in a tree would make the tree die in the field, so camphor trees have higher tolerance to insects' infection. With a sample size of 3 twigs per tree on average only reached 0.9 - 0.4 precision level as density increasing in our investigation.

In low population density, this sampling would have poor precision, though estimated density was 1 scales, there would be 0-2 scales i.e., with 100 percent error. In higher population density, the estimated population would reach required precision; For example, when the mean is 100 scales per twig, its precision level would be 0.5, while the real population would be 100 $\pm$ 25 scales. At heavily damaged density, e.g. 296 scales per twig, this sampling would reach 0.43 precision level and this precison could not influence the decision of control.

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摘 要

許如君[1,2]、洪淑彬[2]、吳文哲[2]* 2001 樟白輪蚧（*Aulacaspis yabunikkei*）在樟樹上之空間分布及取樣技術 植保會刊 43: 69−81.
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比較樟白輪蚧（*Aulacaspis yabunikkei* Kuwana）各齡期在樟樹上株間、枝間及葉間的 Iwao's patchiness regression 及 Taylor's power law 的空間分布指數及決定在樟樹上的取樣單位及最適取樣齡期。結果以 Taylor's power law 的相關性較高（R^2：0.89 - 0.99），Iwao's patchiness regression 則較差（R^2：0.54 - 0.90）。各齡期在株間、枝間及葉間的 Taylor's power law 及 Iwao's patchiness regression 聚集指數範圍分別為 1.76 - 2.65 及 1.26 - 11.83，顯示各齡期在樟樹上的分布都屬聚集型（$b > 1.7$ 及 $\beta > 1.2$）。樟白輪蚧在樟樹上內層的分布顯著較外層多，在老葉上分布較嫩葉上多，且葉背的數目也較葉面上多。利用樟樹上分層取樣的結果，各齡期和總蟲數的相關性以卵為最高（$R^2 = 0.89$，$P < 0.01$），適合推薦專業人士取樣估計總蟲數。不過因卵太小難以肉眼觀察計數，推薦以株內枝條上老葉上的雌介殼數（和總蟲數的相關性，$R^2 = 0.68$，$P < 0.01$）為第一線防治人員取樣的對象。在樟白輪蚧嚴重危害時（每株 100 隻時），如只要求 0.5 的精密水平時，則只需取樣 3 株即具代表性。

(關鍵詞：樟白輪蚧、空間分布、取樣)

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