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E. T. Stratopoulou, E. T. Kapatos

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Distribution of Population of Immature Stages of Pear Psylla, *Cacopsylla pyri*, within the Tree and Development of Sampling Strategy¹

E.T. STRATOPOULOU and E.T. KAPATOS

National Agricultural Research Foundation, Plant Protection Institute of Volos, Greece

ABSTRACT

The distribution of *Cacopsylla pyri* (L.) (Homoptera-Psyllidae) infestation within the tree canopy and in the various organs of the tree was investigated in the region of Magnesia (Greece) during 1988 and 1989. The upper parts of the tree canopy and the sections orientated to south and west were infested heavier during winter and spring. Later in the season the infestation was diffused within the tree canopy and at the end of the season a rather uniform pattern of distribution was observed. During the first oviposition period (February-March), the preferred oviposition sites were flower buds while during spring (second generation) they were the leaves of flower buds and young shoots. Sampling strategy for the study of populations of immature stages of pear psylla is discussed on the basis of these findings.

Introduction

Pear psylla, *Cacopsylla pyri* (L.), (Homoptera: Psyllidae) has received considerable attention the last years due to the great problems created by this insect in almost all pear growing areas in Greece and other countries (Hodkinson 1984, Broumas et al. 1989).

Although many aspects of pear psylla biology have been investigated (Bonnemaison et Missonier 1956, Nguyen 1962, Atger 1979) it was realized that detailed population studies, which will comprise the simultaneous assessment of the main population parameters, are necessary to provide the basis for the development of a pest management system utilizing ecological criteria. Therefore, an intensive study of the population dynamics of *C. pyri* was initiated in the region of Magnesia (eastern-central Greece) in 1988 that included the study of the distribution of pear psylla infestation within the tree, the estimation of development times of the various stages and potential fecundity of adult females under field conditions and

the assessment of the action of mortality factors.

The knowledge of the distribution of the infestation of pear psylla within the tree, as in most insects, is considered necessary for the development of optimum sampling plans (Southwood 1978) but such knowledge is limited (Onillon and Bassino 1976). Additional problems exist, however, in sampling the population of immature stages of this insect. The oviposition sites of females in the first generation are different from those in the other generations. During the first generation pear psylla oviposits, mainly, at the base of flower buds and the young larvae enter the opening flower buds to complete development. In the second generation females oviposit, mainly, on the leaves developed from flower buds, leaf buds and young shoots; the fully developed larvae (L₄₋₅) are usually found at the axil of the leaves (Bonnemaison and Missonier 1956). This behaviour creates problems in the selection of a unique sampling unit for the whole infestation period. Moreover, pear psylla infestation is very high during certain periods of the year (e.g. late spring-early summer) and it is important to keep the number of samples to a minimum.

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Therefore, the pattern of sampling (random or stratified) is critical to minimize variance (Southwood 1978).

For these reasons, it was considered necessary to study the distribution of pear psylla infestation within the tree and in the various organs in order to establish a sampling strategy for the population studies.

Material and Methods

The study was carried out during 1988 and 1989 at an experimental site (Koropi) where the population dynamics of pear psylla has been studied for a number of years (Stratopoulou and Kapatos, unpublished). The trees used for sampling were of the variety «Kristali» and of medium size (4-5 m). They were kept free from insecticidal treatments but they received the usual agricultural practices (pruning, fertilization, etc.).

In order to determine the distribution of pear psylla infestation within the tree canopy samples were taken from two levels (upper and lower part of the canopy) and the four directions (E, S, W, N), dividing thus the tree canopy into eight sections. Four trees were selected for sampling at the beginning of the season and samples were obtained three times in 1988, i.e. on March 22 (first generation), May 18 (second generation) and October 13 (last generation). The samples were taken to the laboratory and examined under a microscope and all stages found were recorded.

In the first sampling, two samples were taken from each section of the four trees (a total of 64 samples) and the sampling unit consisted of eight flower buds. The data were analysed with a factorial anova of mixed model (Sokal and Rohlf, 1969) where Levels and Directions were considered as fixed factors and Trees as random factor and therefore two samples per section are required to obtain an unbiased estimate of the residual error. According to the model, the random factor (Trees) is tested over the residual error but each fixed factor is tested over its interaction with the random factor if this was found significant.

In the second and third samplings, however, one sample per section was taken (a total of 32 samples) and results were analysed with a factorial anova of fixed model (Trees are considered as blocks). This was decided firstly, because infestation at these periods is, usually, high and it would be difficult to examine a great number of samples during a reasonable period of time. Secondly, as the main statistical differences between the various sections of the tree canopy would have been investigated in the first sampling period, it was considered sufficient to examine only whether the main trends of the distribution of infestation determined in the first sampling are still observed during the rest of

the year or the infestation is diffused and a more uniform pattern of distribution is established.

In the second sampling, the sampling unit consisted of the leaves developed from five flower buds while in the third sampling, in autumn, it consisted of the leaves of five leaf buds.

The distribution of pear psylla infestation in the various organs of the tree (flower buds, leaf buds and young shoots) during the year was studied by sampling the various organs three times (March, May, October) in 1988 and 1989. In the first sampling period (March) a number of two year growth small branches was randomly chosen from the same section of six trees. From these branches, 20 flower buds and 20 leaf buds were taken and examined for pear psylla infestation after classifying the flower buds in different stages of development (according to Fleckinger, Bonnemaison 1962). In the second and third sampling periods (May, October) in addition to the flower buds and leaf buds the young shoots were also sampled in the same way as the other organs and the results were expressed both as number of living individuals per organ and per leaf. The results were subjected to an analysis of variance in a randomized block design and Duncan test.

Results

Figure 1 shows the distribution of the population of pear psylla within the tree in relation to Levels (upper and lower part of the tree canopy) and Directions (E, S, W, N) on March 22, 1988. Table 1 gives the statistical significance of the differences due to Trees, Levels and Directions and their interactions found in the analysis of variance which was carried out on these data.

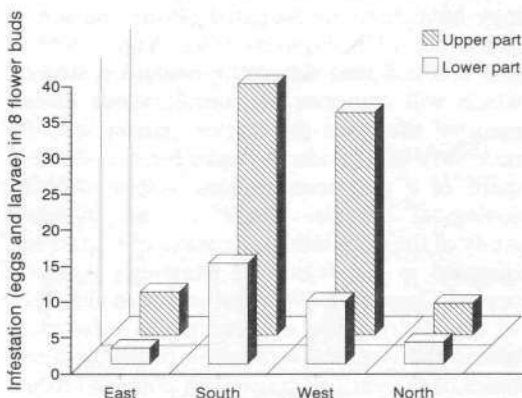


Figure 1. Distribution of pear psylla infestation (number of eggs and larvae in eight flower buds) within the tree canopy in relation to Levels (upper and lower part) and Directions (E, S, W, N) on March 22, 1988.

TABLE 1. Statistical significance of variation due to Trees, Levels, Directions and their interactions in trees infested by pear psylla, on March 22, 1988.

Sources of variation	Statistical significance
Trees	n.s
Levels	n.s
Directions	n.s
Trees \times Levels	*
Trees \times Directions	**
Levels \times Directions	n.s
Trees \times Levels \times Directions	**

** Significant at 0.01 level

* Significant at 0.05 level

n.s Non significant

The results indicate that pear psylla infestation was higher in the upper parts of the tree canopy compared with the lower parts and in the southern and western sections of the tree compared with the eastern and northern sections. The analysis of variance, however, indicated that there were the interactions of Levels and Directions with the random factors (Trees) and also the higher order interaction (Trees \times Levels \times Directions) that were significant. Levels and Directions, despite the great differences observed, were not found significant because according to the model they were not tested over the residual error but over the corresponding interaction with the random factors (Trees), since these were found significant.

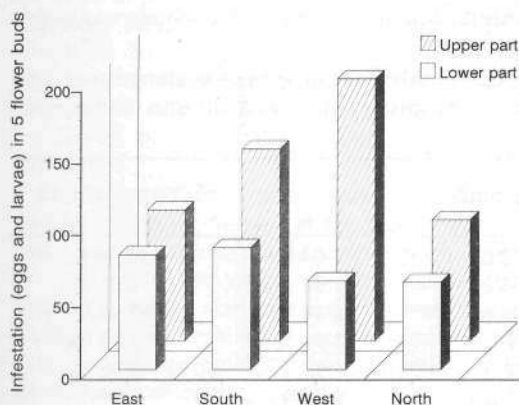


Figure 2. Distribution of pear psylla infestation (number of eggs and larvae on the leaves of five flower buds) within the tree canopy in relation to Levels (upper and lower part) and Directions (E, S, W, N) on May 18, 1988.

TABLE 2. Statistical significance of variation due to Trees, Levels, Directions and their interactions in trees infested by pear psylla, on May 18, 1988.

Sources of variation	Statistical significance
Trees	n.s
Levels	*
Directions	n.s
Trees \times Levels	n.s
Trees \times Directions	n.s
Levels \times Directions	n.s

* Significant at 0.05 level

n.s Non significant

Figure 2 shows the distribution of pear psylla infestation within the tree canopy in samples taken on May 18, 1988 (second generation) and Table 2 gives the statistical significance of the differences resulted from the analysis of these data. The infestation was again higher in the upper parts of the tree canopy and in the southern and western sections but the differences between the various sections were, statistically, much smaller compared with the corresponding differences observed in the first sampling. The analysis of variance carried out on these data indicated that only Levels were significant at the 0.05 level of significance but the comparisons in the F-test were made, for reasons explained in the previous chapter, according to a fixed model of factorial anova

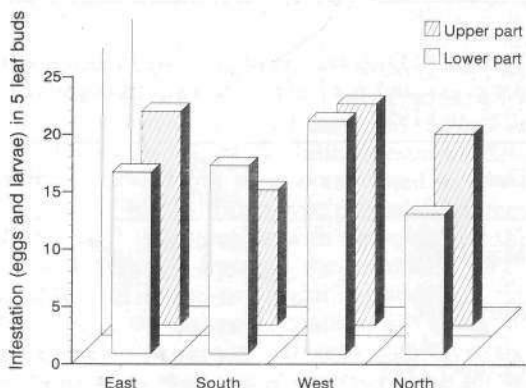


Figure 3. Distribution of pear psylla infestation (number of eggs and larvae on the leaves of five flower buds) within the tree canopy in relation to Levels (upper and lower part) and Directions (E, S, W, N) on October 13, 1988.

(Trees are considered as blocks) i.e. the various components of variance were tested actually over the higher order interaction (Trees x Levels x Directions).

Figure 3 shows the distribution of pear psylla infestation within the tree-canopy in samples taken in October 13, 1988. No substantial differences were observed between the various sections of the tree and this was confirmed by the statistical analysis carried out on these data.

Table 3 gives the distribution of the population of pear psylla (mainly eggs and young larvae) in the various organs of the tree (flower buds and leaf buds) during the first oviposition period in 1988 and 1989. In both years, most of the infestation (95.5% in 1988 and 92.3% in 1989) was found on flower buds.

Figure 4 gives the mean number of eggs per flower bud, which was classified in different stages of development, found in samples taken during the first oviposition period in 1988 (March 14) and 1989 (March 10). The results indicated that the more advanced flower buds (stage D in 1988 and E in 1989) were heavier infested but the differences were not statistically significant even in the 1989 data, due, appar-

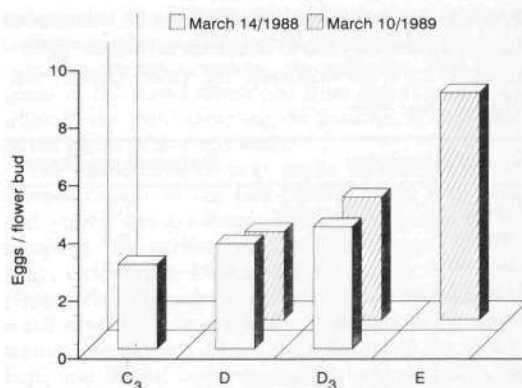


Figure 4. Mean number of eggs per flower bud classified according to its stage of development, on March 14, 1988 and March 10, 1989.

ently, to the high variability of the data.

Table 4 gives the population of immature stages of pear psylla on leaves developed from flower buds, leaf buds and young shoots in two years growth branches during spring (May) and autumn (October) of 1988 and 1989. Results are expressed both as number of individuals per organ and per leaf. According to the results and the statistical significance of the differences, which are given in the same table, during spring flower buds and newly developed shoots were much heavier infested than leaf buds. When the results are expressed as population density of leaves, infestation on the leaves of young shoots is not so high as when results are expressed per organ because the mean number of leaves of young shoots is, usually, much higher than of the other organs. During autumn, however, the infestation on the leaves of leaf buds was higher

TABLE 3. Proportion (%) of the total infestation (eggs and young larvae) in flower buds and leaf buds on March 14, 1988 and March 10, 1989.

Date	Flower buds	Leaf buds
March 14, 1988	95.5	4.5
March 10, 1989	92.3	7.7

TABLE 4. Distribution of pear psylla infestation (mean number of individuals of immature stages per organ and per leaf) in the various organs of the tree in spring (May) and autumn (October) of 1988 and 1989.

Date	Type of estimate	Flower buds	Leaf buds	Young shoots
May 1988	per organ	33.82 c	8.04 a	30.34 bc
	per leaf	7.04 c	1.91 a	4.21 bc
May 1989	per organ	14.14 bc	3.44 a	17.94 c
	per leaf	3.14 c	0.91 a	2.19 bc
October 1988	per organ	1.80 a	7.92 c	3.30 ab
	per leaf	0.39 ab	1.71 c	0.33 a
October 1989	per organ	3.48 a	6.24 a	5.82 a
	per leaf	0.84 a	1.23 a	0.63 a

Means in rows followed by different letter differ significantly at the 0.05 level of significance.

than in the other organs but the differences were statistically significant only in the results of 1988.

Discussion

The analysis of the results obtained in this study indicated that pear psylla infestation is not randomly distributed in relation to the various organs and the various sections of the tree.

During the first oviposition period (February-March) the preferred oviposition sites of the females of *C. pyri* are in the upper parts of the canopy and orientated to south and west, probably because temperature requirements are better met in these sections. This distribution, however, may be influenced by the tree (number of available oviposition sites, position of the tree, etc.) as indicated by the statistical significance of the interactions of both Levels and Directions with the Trees. Previous work in France (Onillon and Bassimo 1976) for the same period indicated the preference of pear psylla females to oviposit on the flower bud bearing branches in the southern and western sections of the tree.

The build up of the population of pear psylla during the second generation causes the diffusion of the infestation within the tree canopy and differences in the infestation between the various sections of the tree become smaller. At the end of the season (October), the distribution of the infestation within the tree in relation to Levels and Directions becomes rather uniform. At this time of the year, however, most of the leaves of unprotected pear trees, particularly in the more heavily infested sections, are in bad condition due to high infestation earlier in the season and, therefore, the main factor that determines the oviposition behaviour of females is, probably, the suitability of oviposition sites.

During the first oviposition period, the females oviposit, mainly, at the base of flower buds. This behaviour maximizes the probability at survival of the young larvae, which after hatching enter the opening flower buds. Further on, the results indicated a tendency for preference of the females to oviposit in the more advanced flower buds although this was not supported statistically and, therefore, it needs further investigation. This behaviour, if it exists, should be based on the same physiological mechanism that causes the attraction of the

females to flower buds and it would further maximize the probability of survival of young larvae because the synchronization of the progressive egg hatching and the progressive opening of flower buds would be more successful.

During the second generation, the females of pear psylla show a preference to oviposit on the leaves of young shoots, that probably provide a better substrate for the development of the larvae, and on the leaves of flower buds probably because most of the adults of the first generation are produced in flower buds bearing branches.

Later in the season (October), it appears that leaves developed from leaf buds are the preferable oviposition sites but this could be the result of the bad condition of the leaves of the other organs after they have been heavily infested earlier in the season.

The results and the conclusions from this study provide the basis for the development of a sampling programme for the study of pear psylla populations that will minimize «systematic» errors (LeRoux and Reimer 1959) which arise from a random sampling in a non randomly distributed population. It is suggested, therefore, that samples for the estimation of pear psylla population of immature stages must be taken from the four directions of the tree particularly during winter and spring and from the middle of the tree canopy that expresses the general trend of the distribution of population in regard to height.

Because the oviposition sites of the females in the first generation are different from those in the other generations, the sampling unit for the whole infestation period should be composed of a constant number of organs rather than a constant number of leaves. However, as the number of leaves of the various organs, after they have been developed and until the end of the season, is rather constant, infestation data for this period can be also expressed as infestation per leaf for other type of studies or even connect these estimates with those during the first generation through the estimated mean number of leaves per organ. In addition, the sampling unit should contain all kinds of organs proportionally to their availability in the tree in order to obtain a weighted mean of the infestation of each section (Cochran 1963).

Undoubtedly, the size of the sampling unit and the number of samples required to attain a certain precision are important parameters in

developing optimum sampling plans. However, as these parameters often are a function of population density (Southwood 1978) and they are, usually, determined by cost they should be examined specifically in each particular case.

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KEY WORDS: *Cacopsylla pyri*, Distribution of *Cacopsylla pyri*, Sampling of *Cacopsylla pyri*.

Κατανομή του Πληθυσμού των Ατελών Σταδίων της Ψύλλας της Αχλαδιάς (*Cacopsylla pyri*) στο Δένδρο και Ανάπτυξη Στρατηγικής Δειγματοληψίας

Ε.Θ. ΣΤΡΑΤΟΠΟΥΛΟΥ και Ε.Θ. ΚΑΠΑΤΟΣ

Εθνικό Ίδρυμα Αγροτικής Έρευνας
Ινστιτούτο Προστασίας Φυτών Βόλου

ΠΕΡΙΛΗΨΗ

Η κατανομή του πληθυσμού των ατελών σταδίων της ψύλλας της αχλαδιάς (*Cacopsylla pyri*) στο δένδρο μελετήθηκε στην περιοχή της Μαγνησίας στη διάρκεια των ετών 1988 και 1989. Τα υψηλότερα τμήματα του δένδρου καθώς και τα τμήματα που είναι προσανατολισμένα στο Νότο και στη Δύση βρέθηκαν να είναι περισσότερο προσβεβλημένα στη διάρκεια της πρώτης και δεύτερης γενιάς. Αργότερα, οι διαφορές εξομαλύνονται και στο τέλος της περιόδου η κατανομή γίνεται σχεδόν ομοιόμορφη. Στη διάρκεια της πρώτης περιόδου ωοτοκίας (Φεβρουάριο-Μάρτιο) τα θηλυκά γεννούν τ' αυγά τους κατά προτίμηση στη βάση των καρποφόρων οφθαλμών και πολύ λιγότερο στους φυλλοφόρους οφθαλμούς και μάλιστα παρατηρήθηκε η τάση να γεννούν περισσότερα αυγά στους πιο ανεπτυγμένους οφθαλμούς. Στη διάρκεια της δεύτερης γενιάς περισσότερα αυγά εναποτίθενται στα φύλλα των ετήσιων βλαστών και καρποφόρων οφθαλμών απ' ότι στα φύλλα των φυλλοφόρων οφθαλμών. Το φθινόπωρο όμως παρατηρείται το αντίθετο φαινόμενο. Με βάση τα παραπάνω, αναπτύσσεται η στρατηγική της δειγματοληψίας που πρέπει να ακολουθείται για τη μελέτη του πληθυσμού του εντόμου αυτού.