### Estimating the Density of Beech Caterpillar, *Quadricalcarifera punctatella*, by Frass Drop Samples

#### Naoto KAMATA, Yutaka IGARASHI, Toshio YANBE\* and Masatoshi IGARASHI\*\*

Tohoku Research Center, Forestry and Forest Products Research Institute (Shimokuriyagawa, Morioka, Iwate, 020-01 Japan)

#### Abstract

The purpose of this study was to investigate frass production of the beech caterpillar, Quadricalcarifera punctatella (Motschulsky) (Lep., Notodontidae) and to develop a method for estimating the field density of its last instar larvae from fallen pellets. About 90% of larval frass by weight was produced during the last instar period. The number of pellets falling to the forest floor was a better parameter for density estimation than weight. Pellets with a width of more than 1.8 mm were considered to have been produced during the last instar. The number of pellets produced per day or per hour in the last instar was influenced by many factors, especially by temperature; however, the number of pellets produced during the last instar was relatively constant (ca. 630) regardless of temperature and photoperiod. The number of last instar pellets dropping per m<sup>2</sup> during the season was measured in traps and divided by 630 to estimate the density of the last instars. This method, based on the Southwood-Jepson graphical method, is very simple but reliable because the estimates are not influenced by ambient temperature, and because the total number of falling pellets can be estimated correctly by setting traps throughout the entire season. Although some frass was destroyed and washed away from the trap, we concluded that no compensation was necessary for field application since the decrease was minimal. The density was underestimated when severe defoliation occurred. In such a situation, the density (Y) should be estimated from the following equation: Y =0.988110g10 X + 0.898, where X is the maxium value of the frass drop per day (g. dw/m²/day). Only five traps (1 m² each) were required with a 0.2 ratio of tolerance limit at the 70% confidence level. Such a small number of samples were necessary because the method used counts of frass pellets rather than individuals; larval mobility and wind homogenized the spatial distribution of falling frass.

Discipline: Insect pest/Experimental apparatus and method Additional key words: forest defoliator, frass trap, Insecta, sample size

Present address: \* Forestry Development Technological Institute (Sugo 208-40, Takizawa, Iwate, 020-01 Japan)

\*\* Yashima Co., Ltd. (Orimoto 540, Shimodate, Ibaraki, 308 Japan)

#### Introduction

The beech, *Fagus crenata* Blume, is a dominant species in the cool temperate forests in Japan. The beech caterpillar, *Quadrical-carifera punctatella*, feeds on beech leaves, sometimes defoliating the trees completely. However, there are few population studies except for some short reports on the insect's life history and some conspicuous phenomena during outbreaks<sup>2)</sup>.

It is difficult to count directly the number of insects living in the forest. Frass drop has been used as an index of both population density and insect damage by many forest entomologists. In a population study, especially on population dynamics between generations, the total number or generation density is as important as seasonal occurrence. There are several different statistical methods for estimating the number of individuals entering a stage or at the mid-point of a stage.

In this paper, we report a method of estimating the last instar larval density of Q. punctatella from fallen frass pellets in beech forests. We then discuss several problems relating to field surveys and the number of traps necessary to obtain a density estimate at a given level of precision.

### Frass production of larvae<sup>4)</sup>

Rearing experiments showed that the last instar larvae produce 85-92% (mean = 90.3%) of the total frass; defoliation caused by *Q*. *punctatella* is due primarily to this stage. Since body size affects the weight of frass but not the number of pellets, the number of frass pellets is a more appropriate measurement for estimation of larval density than frass weight. Pellets larger than 1.8 mm in width were considered to have been produced by last instar larvae.

The daily frass production was greatly in-

fluenced by the age (days old) after entering the last instar. The number of pellets produced at 5 days (mean = 106.7, S.D. = 12.2) was about three and a half times that at the start of the instar (mean = 32.4, S.D. = 24.3). The number of pellets produced per hour also varied widely





Larvae were reared under constant conditions (14L-10D and 70% RH regime). a: Number of pellets produced per day

- during the last instar.
- b: Total number of pellets produced during the last instar.



Fig. 2. Influence of photoperiod on the total number of pellets produced by the last instar larvae of Q. punctatella (Mean  $\pm$  S.D.)

Larvae were reared under constant conditions at 21°C and 70% RH.

among individuals. Fig. 1 shows the influence of temperature on frass pellet production (14L-10D hr, 70% RH.); the number of pellets produced per day was greatly influenced by temperature. However, the total number of frass pellets produced during the last instar was almost the same regardless of temperature, and variations among individuals were small (C.V.=0.07-0.14). The total number of pellets produced during the last instar was not influenced by photoperiod with only one exception: under the 16L-8D regime, the average number of pellets was higher (Fig. 2).

Since the number of pellets produced daily was greatly influenced by age and temperature, this factor was not considered as a good parameter for density estimation in the field. On the other hand, since the total number of frass pellets produced during the last instar was almost constant regardless of temperature and photoperiod, this factor was considered to be a reliable parameter for estimating the density of larvae.

### Method of density estimation from falling frass<sup>4)</sup>

### 1) Modification of the Southwood-Jepson graphical method

The total density of the last instar in the season is then calculated using the following equation:

 $Density = \frac{Total no. of last instar}{Total no. of pellets per m^2}$ Total no. of pellets produced per individual during the last instar

..... (1)

### 2) Total number of pellets used for the density estimation

The last instar larvae occur from mid-July to late August in the northern part of the Tohoku district. Daylight hours in Morioka city (141°09'E, 39°42'N) decreased gradually from 14:49 (Jul. 10) to 13:12 (Aug. 29)<sup>6)</sup>. Since light changes gradually in the field, the switching point of dark and light for *Q. punc*-*tatella* cannot be determined exactly. The moth produces only one generation a year, so that photoperiod was shorter than the critical photoperiod (15.5–16 hr)<sup>2)</sup>. Hence, the mean value averaged over 4 regimes of different temperatures under a 14L–10D hr photoperiod (630) should be used for density estimation as the best estimate of the total number of pellets produced during the last instar.

### 3) Advantages and disadvantages of the method

This method is based on the Southwood-Jepson graphical method (SJ in this paper), which is one of the techniques for estimating the number of stages from a series of population samples<sup>70</sup>. Though SJ is a simple method for integration under a curve, it has many disadvantages described below. The technique described in this paper solves the following problems of SJ and methods for estimating larval density from falling frass (FF in this paper): (1) Estimates are not influenced by temperature; the parameter used for the total number of nellets produced during the last instar is

of pellets produced during the last instar is constant regardless of temperature (Problem common to SJ and FF).

(2) If frass-traps are set continuously throughout the season and falling pellets are collected frequently, the total number of pellets produced by the population during the season can be estimated accurately without daily checking (Problem of SJ).

(3) Continuous frass-traps can collect some portions of the pellets remaining on the foliage (Problem of FF).

(4) The total number of total density throughout the generation must be determined rather than only a proportion of the total generation (Problem of FF). However, there are some potential disadvantages:

 Some pellets are destroyed, decomposed or washed away by rainfall (Problem of FF).
 A change in survivorship alters the age of the stage to which the estimate of the population relates (Problem of SJ)<sup>3)</sup>.

## How to deal with the problems in field surveys<sup>3)</sup>

### 1) Frass destroyed and washed away by rainfall

The frass pellets produced by the last instar larvae of Q. punctatella were left for 15 days in a rectangular trap  $(1 \times 1 \text{ m})$  made of cloth and exposed to the elements. The dry weight of frass decreased by 0.068 g (1.9%), the difference being due to the removal of frass by rainfall (Table 1). The decrease in the number of pellets was less (3.3%) than the decrease in weight (5.9%) because most pellets had not been destroyed but had become smaller in size. The mean weight of a pellet decreased from  $4.334 \times 10^{-4}$ g (97.3%) during the experiment. If the pellet is considered to be a sphere, then the radius should decrease to 99.1% of the original value. The width threshold of last instar pellets decreased to 1.78 mm from the original value of 1.80 mm.

The degree of pellet destruction is likely to

 
 Table 1. Amount of frass of Q. punctatella destroyed and washed away from a trap

		Before exposure	After exposure	
Pellets	(g•dw) (No.)	3.670 (8,468)	3.452 (8,185)	
Powder (g.dw)		-	0.150	
Total	(g•dw)	3.670	3.602	

The last instar frass was exposed to the elements in a trap for 15 days. The number of frass pellets and the dry weight of the pellets and powder were determined before and after exposure. change in response to many factors such as precipitation, weather, wind, vegetation on the forest floor, trap design, and amount of frass in the trap. The amount of destroyed and/or washed away frass was negligible, although two typhoons and abundant precipitation occurred during this experiment. We concluded that no compensation was necessary for the loss associated with destroyed and washed away frass.

### 2) Supplemental method applied in an outbreak period

The cumulative frass drop of last instar larvae is usually determined by the larval density. However, when the larval density is extremely high and all foliage is defoliated, the biomass of the canopy determines the level. Thereafter we also tried to estimate the larval density from maximum daily frass drop rather than from the cumulative level. The maximum value of successive measurements (X) expressed on a logarithmic scale gave the best correlation with larval density (Y). The regression line was given by;

$$\log_{10} Y = 0.988 \log_{10} X + 0.898 \quad (r = 0.991)$$
.....(2)

When all beech trees were defoliated, density estimates could be calculated by equation (2). This method, however, could not be used under conditions of extremely high density when all beech trees are defoliated before frass drop reaches the maximum level. The head capsule may be a better method for density extimation under extremely high densities<sup>1)</sup>, though many more traps would be necessary for low density populations.

# Field applications and necessary sample size<sup>3)</sup>

### 1) Collecting frass pellets in fields

Cloth-made traps (1 m<sup>2</sup> each) were deployed near the forest floor in each study plot ( $20 \times 20$  m) from mid-July to late August (Plate 1). The trap contents were brought back to the laboratory at intervals and dried in an electric drier at 60°C. The frass was separated from the other debris, then sorted using a sieve and by hand into three categories: frass pellets of the last instar larvae of Q. punctatella, other



Plate 1. Frass traps deployed on a forest floor

pellets, and powdered frass. Powdered frass was divided by weight and added to the other two categories in proportion to weight. The number of last instar pellets of *Q. punctatella* was then calculated from the weight. The total density was estimated from equation (1), and from equation (2) when severe defoliation occurred.

### 2) Tolerance limit of estimation and required sample size

When CV is the coefficient of variance, and *E* is the tolerance limit (= (m-M)/m (*m*: the mean value of samples, *M*: the mean value of the whole population)), the necessary number of samples (*n*) is given by;

$$n > = t_0 C V^2 / E^2$$
, .....(3)

where  $t_0$  is a critical value of Student's distribution for the required confidence level<sup>3)</sup>.

Twenty traps were deployed in two plots in Appi, Iwate Pref. during 2 years, 1987 and 1988. Variances in the number of frass drop are shown in Table 2. We investigated the number of traps necessary to maintain tolerance limits E of 0.1, 0.15 and 0.2 for a 70% confidence level ( $t_0 = 1$ ). When drop was established from five traps, the ratio was larger than 0.15 but smaller than 0.2 in one plot where frass drop varied the most. The relationship between larval density and its coefficient of variance was investigated using five traps in 15

Table 2. Necessary number of traps to obtain a given level of precision (confidence level = 0.7)

Plot no.	Year	Density (/m <sup>2</sup> )	CV	Ratio of tolerance limit		
				1	1987	1.79
1988	3.19	0.227	6		3	2
3	1987	0.634	0.200	4	2	1
	1988	1.15	0.444	20	9	5

Last instar larval density and coefficient of variance (CV) in *Q. punctatella* was investigated in Appi Highlands using frass pellets collected from 20 traps in each plot.



Fig. 3. Relation between estimates of last instar larval density and CV (left), and ratio of tolerance limit given by 5 traps at the 70% confidence level (right)

plots of 3 different areas (Fig. 3). The variance decreased with increasing density. Ratio of the tolerance limit was less than 0.2 when the density exceeded  $0.1/m^2$ . These results suggest that five traps are adequate for estimating larval density when the density is higher than  $0.1/m^2$ .

### Conclusion

It has been reported that the number and weight of frass pellets produced over time are strongly influenced by the temperature<sup>4)</sup>. However, it was observed that in many Lepidopteran species the number of pellets produced during any given stage and/or during the entire larval stage was constant regardless of temperature and other factors. The method of density estimation developed in this paper would be equally applicable to these species. If the horizontal area of traps is constant, many small traps can provide better estimates<sup>5)</sup>. This is especially important for short-term investigations. Fewer samples are needed in this technique because frass pellets are sampled rather than individuals. A mean of 630 frass pellets are produced during the last instar stage. The last instar stage lasts 10-15 days under field conditions and larvae move frequently. The direction and intensity of wind are typically variable. All of these factors randomize the spatial distribution of frass drops and enable precise estimation with a relatively small number of large traps.

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