Observations on the feeding behaviour of late-instar larvae of *Choristoneura fumiferana*

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Abstract—Laboratory observations revealed that late-instar larvae of the eastern spruce budworm (*Choristoneura fumiferana* (Clemens)) (Lepidoptera: Tortricidae) spend most of their time spinning, wandering, and resting; less than 10% is spent feeding. Larvae feed in a discontinuous pattern of short feeding bouts separated by much longer intervals of nonfeeding activity. Over a 2 h observation period, feeding bouts averaged 2.2 min and were separated by 17.4 min intervals for 4th-instar larvae as compared to 3.3 min bouts separated by 33.4 min intervals for 5th-instar larvae. The duration of a feeding bout was positively correlated with the duration of the subsequent interval, not with the duration of preceding intervals, suggesting that feeding-bout frequency is governed primarily by post-ingestion processes. It is postulated that short feeding bouts followed by long intervals limit the window for ingesting an efficacious dose of aerially applied insecticides such as *Bacillus thuringiensis*.

Résumé—Des observations en laboratoire révèlent que les larves des derniers stades de la tordeuse des bourgeons de l'épinette de l'est (*Choristoneura fumiferana* (Clemens)) (Lepidoptera : Tortricidae) passent la majorité de leur temps à tisser, errer et se reposer; moins de 10 % du temps est utilisé pour l'alimentation. Les larves se nourrissent en suivant un patron discontinu de courtes périodes d'alimentation séparées par des intervalles beaucoup plus longs sans activité alimentaire. Sur une période d'observation de 2 h, les périodes d'alimentation durent en moyenne 2,2 min et sont séparées par des intervalles de 17,4 min au 4e stade et durent 3,3 min avec des intervalles de 33,4 min au 5e stade. Il existe une corrélation entre la durée de la période d'alimentation et la durée de l'intervalle subséquent, mais non avec la durée de l'intervalle précédent; cela laisse croire que la fréquence des activités alimentaires est contrôlée principalement par les processus postérieurs à l'ingestion. Nous avançons l'hypothèse selon laquelle les courtes périodes d'alimentation suivies de longs intervalles peuvent restreindre la fenêtre d'ingestion d'une dose efficace des insecticides répandus par avion, tels que *Bacillus thuringiensis*.

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Foliage-feeding Lepidoptera spend most of their larval life on host foliage and are readily viewed as feeding continuously (*e.g.*, Heinrich 1971; Truman 1972). The feeding patterns of relatively few species of folivorous insects have been analysed in detail. Continuous feeding, interrupted only by moulting or changes in feeding position, has been reported for some sawfly species (Heitland and Pschorn-Walcher 1993), whereas feeding by *Locusta migratoria* L. (Orthoptera: Acrididae), *Manduca sexta* (L.) (Lepidoptera: Sphingidae), and *Pieris brassicae*

L. (Lepidoptera: Pieridae) is characterized by relatively short episodes of active feeding (hereinafter referred to as bouts) separated by extended resting periods (bout intervals) (Ma 1972; Simpson 1982; Reynolds *et al.* 1986; Bowdan 1988). Larvae of the eastern spruce budworm, *Choristoneura fumiferana* (Clemens) (Lepidoptera: Tortricidae), also have a pattern of discontinuous feeding, but we are unaware of any published studies on temporal aspects of its feeding behaviour. We examined spruce budworm feeding behaviour under laboratory

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Table 1. Distribution of time (% of total) spent on various activities by late-instar larvae of the eastern spruce budworm (*Choristoneura fumiferana*) over a 2 h observation period.

Instar	n	Feeding	Wandering	Spinning	Resting
4	39	$10.0 \pm 1.0a$	$7.0 \pm 1.5a$	$31.0 \pm 2.4a$	$52.0 \pm 3.8a$
5	27	$8.2 \pm 0.8a$	$13.9 \pm 1.9ab$	$28.4 \pm 2.4a$	$49.5 \pm 2.8a$
6	36	$8.3 \pm 0.6a$	$16.5 \pm 1.9b$	$43.6 \pm 3.0b$	$31.6 \pm 2.7b$

Note: Values are given as the mean $\pm$ SE. Within a column, values followed by a different letter are significantly different (Tukey's test, $P < 0.05$).

conditions by comparing time budgets of 4th-, 5th-, and 6th-instar larvae. Feeding bouts and bout intervals were quantified for 4th- and 5th-instar larvae only.

Larvae were obtained from a disease-free colony maintained at the Great lakes Forestry Centre (Natural Resources Canada, Sault Ste. Marie, Ontario) (van Frankenhuyzen *et al*. 2004) and reared on artificial diet. Larvae that had moulted within the preceding 24 h were placed individually into cork-stoppered 0.5-dram vials. A 1.5 cm long new-growth needle of balsam fir (*Abies balsamea* L., Pinaceae) was inserted into a slit in the bottom of each stopper. Positioning the needle in the slit facilitated observation but was not necessary for larval feeding. Five vials were placed in a Petri dish under a dissecting scope at low magnification and the dish was slowly rotated to track behaviours of individual larvae for a period of at least 2 h. Feeding (mandibular movement), wandering (movement off the needle), spinning (silk production), and resting (no discernible movement) were differentiated as key activities. The duration of each activity was recorded for 6–10 groups of 5 larvae each. By rotating the dish, each larva was observed every 10 s. Time spent on each activity was accumulated over the observation period and expressed as a percentage of total observation time.

Each larva typically wandered through its vial before establishing a feeding position along the needle's edge. The observation period for each larva started with the first feeding bout. Feeding was executed in relatively short, distinct bouts consisting of repeated sequences of bites (chewing) directed along the needle's edge in a downward arc, thereby creating a semicircular hole extending across the width of the needle.

Feeding bouts were separated by relatively long periods of nonfeeding activity during which the larvae typically moved off the needles and retreated into a silk shelter spun against the sides or bottom of the vial, in which they would rest or spin until the next bout.

Compilation of the various recorded activities into an overall time budget showed that larvae spent most (90% or more) of the total observation period wandering, resting, and spinning (Table 1). The proportion of time allocated to these activities varied significantly among instars (analysis of variance, General Linear Model, Minitab version 15; $F > 12.4$, df = 2, 101, $P < 0.001$), with 6th instars spending more time spinning and less time resting than did earlier instars. Feeding occupied 10% or less of time regardless of developmental stage ($F = 0.01$, $P = 0.941$) (Table 1).

The duration of each bout and bout interval over the observation period (approximately 2 h) was recorded for each 4th- or 5th-instar larva. Data regarding individual bouts for 6th instars were inadvertently lost after overall time budgets were constructed. Determining the onset and termination of feeding bouts was confounded by the occurrence of short activity gaps within bouts. The graphical method of Bowdan (1988) indicated that a minimum feeding-bout duration of 2 min could be used to separate intrabout gaps from interbout intervals. Thus, a feeding bout was defined as an episode of feeding that was separated from any other such episode by more than 2 min. Similar gap estimates have been reported for *M. sexta* (Reynolds *et al*. 1986; Bowdan 1988). Durations of feeding bouts and bout intervals (Table 2) were not normally distributed for either instar (Anderson−Daring normality test, $P < 0.04$) but the distributions did not deviate far from normality because the mean

Table 2. Duration of feeding bouts and intervals between feeding bouts for 4th- and 5th-instar larvae of the eastern spruce budworm (*Choristoneura fumiferana*).

Instar	Variable	n	Duration (min)				
			Median	Mean	SD	Min.	Max.
4	Bout	176	2.1	2.2	1.0	0.5	7.1
	Interval	138	15.9	17.4	9.4	2.7	47.7
5	Bout	76	3.2	3.3	1.6	0.5	10.9
	Interval	49	32.5	33.8	17.7	7.2	74.4

Fig. 1. Relationship between the duration of the first feeding bout and the duration of the interval following that bout for 5th-instar larvae of the eastern spruce budworm (*Choristoneura fumiferana*).
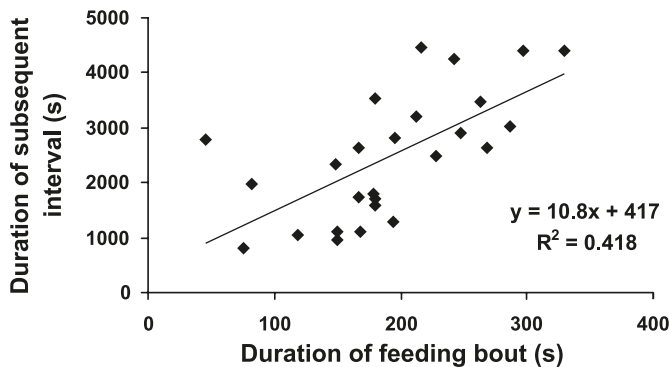
values corresponded closely to the median values. Neither bout duration nor interval duration was significantly affected by their sequence in the case of either instar ($F \leq 2.37$, $P > 0.074$). Bout duration ranged from 0.5 to 10.9 min and was significantly less for 4th instars than for 5th instars (mean: 2.2 *vs.* 3.3 min; $P < 0.001$, df = 103, t = −5.52, two-sample *t* test). Feeding bouts were separated by intervals that ranged from 2.7 to 74 min, with 5th instars exhibiting significantly longer intervals than did 4th instars (mean: 33.8 *vs.* 17.5 min; $P < 0.001$, df = 58, t = −6.22, two-sample *t* test). Thus, as spruce budworm larvae increase in size, daily food consumption increases proportionally (Retnakaran 1983) through an increase in feeding-bout duration. This, in combination with increased bite size (because of larger mouthparts) and possibly increased bite frequency (Bowdan 1988), results in larger meals, which in turn require more time for post-ingestion processing, leading to longer bout intervals. The strategy of increasing the length but not the frequency of meals during growth has

also been reported for *M. sexta* (Bowdan 1988) and *L. migratoria* (Simpson 1982).

This implies that interval duration, and therefore bout frequency, are governed by post-ingestion food processing rather than by pre-ingestion cues such as "hunger." This is supported by a general lack of correlation between the duration of a feeding bout and the preceding interval for either instar (Pearson's correlation coefficient, 4th instar: $r = 0.051$, $n = 137$; 5th instar: $r = 0.163$, $n = 49$; $P > 0.263$), but a significant positive correlation between the duration of the feeding bout and the subsequent interval, at least for 5th instars ($r = 0.392$, $n = 49$, $P = 0.005$). Such a relationship was not found for 4th instars ($r = 0.108$, $n = 138$, $P = 0.207$), suggesting that mechanisms regulating food intake could vary between instars. For 5th instars the strongest relationship was found between the durations of the first feeding bout and the subsequent interval (Fig. 1; $F_{[1,25]} = 17.24$, $P < 0.001$). The slope of that regression confirms our

observation, based on time budgets, that 5th-instar larvae spent only approximately 10% of their time feeding.

Our observations were conducted under highly artificial conditions over a period that represented less than 10% of the daily feeding cycle, and we wondered whether our results are representative and can be extrapolated to the field. Extensive data on the feeding behaviour of individual "wild" spruce budworm in balsam fir trees in New Brunswick, collected during the late 1980s and early 1990s using video cameras (Nigam 1995), supported our limited laboratory observations. Details of those field studies have not been published, but Nigam (1995) reported that "microhabitat cleaning, excretion and movements within microhabitat occupy more time than spinning activity, and feeding activity is of short duration compared with the other two." From 16 to 27 June 1991, 210 h of recording (day and night) yielded 121 h of observations of larval behaviour. Preliminary analysis revealed that late larval instars spent 7.8% of that time feeding, 41.1% resting, 37.5% spinning, and 13.6% doing other things (S.E. Holmes, Atlantic Forestry Centre, Canadian Forest Service, Fredericton, New Brunswick, personal communication). Spruce budworm feeding behaviour appears to be robust: observations at three different scales (first 20 min (Fig. 1) and first 2 h (Table 1) in the laboratory and 10 days of field observations) yielded comparable estimates of the proportion of time spent feeding.

Detailed knowledge of larval feeding behaviour is useful for increasing our understanding of how larvae interact with aerially applied pesticides. For *Bacillus thuringiensis*, the documented patterns of larval feeding explain why high product potency promotes field efficacy (Bauce *et al.* 2004). Feeding occurs in discrete, widely spaced bouts (Table 1) and dose transfer occurs primarily through consumption of needles that have intercepted small spray droplets. Considering the duration of the interval between feeding bouts and the rapid onset of gut paralysis after pesticide ingestion (Heimpel and Angus 1959), feeding inhibition is, in most cases, initiated during the bout interval that follows dose ingestion.

Thus, the fate of a larva feeding on a needle carrying spray deposits could be determined by the dose it ingests within one particular feeding bout. Feeding inhibition is permanent if the ingested dose is lethal, and can last from hours to days when the ingested dose is sublethal, depending on dose and temperature (van Frankenhuyzen and Nystrom 1987). When cycles of feeding inhibition and recovery are ongoing, the probability of acquiring a lethal dose declines because spray deposits degrade (van Frankenhuyzen and Nystrom 1989). It is thus expected that short feeding bouts followed by long intervals limit the window for successful dose acquisition. For optimum efficacy, droplet potency must be high enough that a single ingested droplet will deliver a lethal dose (van Frankenhuyzen and Payne 1993).

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