Phenology of *Lacanobia subjuncta* (Lepidoptera: Noctuidae) in Washington and Oregon Apple Orchards

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ABSTRACT The phenology of *Lacanobia subjuncta* (Grote & Robinson) (Lepidoptera: Noctuidae) was investigated in 30 apple orchards in central Washington state and northeastern Oregon from 1998 to 2001 (57 total orchard-yr). Adult captures in pheromone-baited traps were fit to a Weibull distribution to model emergence of the first and second generations. Initial capture of first generation adults was observed at 216.2 ± 2.6 degree-days (DD) (mean ± SEM) from 1 March by using a base temperature of 6.7°C. The model predicted that flight was 5 and 95% complete by 240 and 700 degree-days (DD), respectively. Monitoring of oviposition and hatch was used to establish a protandry plus preoviposition degree-day requirement of 160.0 ± 7.7 DD, as well as to provide data to describe the entire hatch period. Egg hatch was 5 and 95% complete by 395 and 630 DD, respectively. The start of the second flight was observed at 1217.1 ± 8.3 DD by using an upper threshold for development of 32°C and a horizontal cutoff. The model indicated that the second flight was 5 and 95% complete by 1220 and 1690 DD, respectively. Second generation hatch was 5 and 95% complete by 1440 and 1740 DD, respectively. A discussion of the potential uses of these detailed phenology data in optimizing management strategies is presented.

KEY WORDS *Lacanobia subjuncta*, degree-days, phenology model, pest management, Weibull distribution

Lacanobia subjuncta (Grote & Robinson) (Lepidoptera: Noctuidae) occurs throughout North America and feeds on a variety of plants (Landolt 1997, McCabe 1980), including row crops, shrubs, trees, and several weed species (e.g., dandelion, bindweed, and mallow). In the mid-1990s, larvae of this insect became a pest of apples in central Washington state and parts of northeastern Oregon (Brunner and Doerr 2000).

Insecticides are the primary means of control for *L. subjuncta.* Although general information on the phenology of *L. subjuncta* is available, there is a lack of sufficient detail or predictive capabilities for making integrated pest management (IPM) decisions. Landolt (1998) reported two generations on apples in Washington. He also reported that *L. subjuncta* overwinter as pupae in the soil and emerge as adults in May and June. Larvae of the first generation are present from early June through July. A second adult flight occurs in July and August with larvae present in August and September. Landolt and Smithhisler (1998) discovered that adult activity could be monitored with the use of a sex pheromone; however, our experience has shown that trap captures do not provide a reliable method of timing insecticides. A random selection of 20 orchards from 2000 to 2001 studied for this current manuscript indicated the number of days from initial adult capture to an optimal spray timing (presented in this study) averaged 44 d but ranged from 34 to 57 d. We feel this variation can be explained by differences in temperature accumulations between sites and years. Knowledge of the larval-age distribution in the field is important for timing of sampling and insecticide applications for management of *L. subjuncta* (Doerr et al. 2004). Visual monitoring of larvae is labor-intensive and also has proven to be a difficult method for timing insecticide applications. To optimize insecticide efficacy and minimize nontarget impacts, a more precise determination of *L. subjuncta* phenology is necessary.

Doerr et al. (2002) reported lower temperature thresholds for egg, larval, and pupal development and estimated degree-days required to complete each immature life stage. However, these data alone were not sufficient to develop phenology predictions that described *L. subjuncta* development through the entire growing season. Specifically, information is needed on emergence of first and second generation adults, degree-day requirements for any protandry and preoviposition period, and first and second generation egg hatch periods. Doerr et al. (2004) explained the importance of being able to predict a time after the majority of egg hatch was complete but before the oldest larvae reach the fourth instar. They presented data that suggested this would be an optimal time for applying insecticide inputs that could maximize control while reducing the need for further applications. In this article, we develop a predictive model for first and second generation adult flight periods, estimate the protandry plus preoviposition requirement, and describe both egg hatch periods. Data from intensive field sampling of larvae are presented along with a discussion that explains how this information will be useful in optimizing management strategies, especially timing of insecticide applications.

Materials and Methods

Adult flight activity was monitored by placing one multipurpose bucket style trap (Unitraps, Phero Tech, Inc., Delta, British Columbia, Canada) in each of 30 apple orchards in central Washington and northeastern Oregon from 1998 to 2001 (57 total orchard-yr). Traps were baited with an *L. subjuncta* sex pheromone lure (Peter Landolt, USDA–ARS, Wapato, WA), and a 12.5-cm^2 Vaportape II insecticidal strip (Hercon Environmental Co., Emigsville, PA) was placed in the bottom of each trap to prevent moth escape. Lures were replaced at 6-wk intervals. Traps in each orchard were monitored once per week from full bloom in apple, *Malus domestica* Borkhausen 'Delicious', until late summer of each year, and the number of adult *L. subjuncta* males was recorded.

In 2000–2001, all immature life stages of *L. subjuncta* were monitored in six orchards from central Washington with high population levels (12 orchard-yr). The relative abundance of each life stage (eggs and L1–L6 instars) was determined by sampling orchard trees and ground cover weeds at regular intervals, described in detail below, throughout the growing season. Samples were generally collected three times per week, with one or two orchards visited per day.

Oviposition and egg hatch were monitored by examining the underside of apple leaves. Fresh egg masses were flagged and monitored daily for hatch. Orchard sampling for egg masses was set at two person-hours per day (total for all orchards visited on that day), and sampling was conducted three times per week (6 person-h per wk). Seventy-nine egg masses were flagged during the first oviposition period in 2000. The difference between initial adult male captures and the first observed egg mass at each orchard was considered the preoviposition period, which included any protandry. During the second generation, detection of unhatched egg masses was difficult (only seven total egg masses from four orchards) because eggs were laid lower in the trees, on alternate hosts in the orchard ground cover, or both. However, once the eggs hatched, neonates and their associated feeding damage were relatively easy to locate. Therefore, the second generation hatch period was estimated based on the relative abundance of first instars during larval sampling. Direct sampling of egg masses was not conducted in 2001 because of the low numbers found in 2000.

Larvae were sampled by visual inspection of host plants, limb jarring, and shaking weed hosts into a bucket. To optimize collection of larvae, sampling was conducted in a targeted, nonsystematic manner rather than a random sample or structured sampling protocol. Targeted sampling of trees and weed hosts was conducted on plants showing recent damage typical of *L. subjuncta* feeding. Visual inspection was done at each orchard to locate areas of larval infestation. This initial screening was done for 15–30 min, and any larvae located on trees or weeds during this inspection were collected. Trees with recent feeding damage were then sampled with a limb-jarring method (Burts and Retan 1973). A beating tray (45 by 45 cm) was held under a tree limb showing feeding damage, and the limb was struck three times with a stiff rubber hose. Dislodged larvae that landed on the tray were collected. Approximately 25–50 trees were sampled by limb jarring at each orchard visit. Once a tree was selected for limb-jar sampling, several limbs were sampled to ensure that larvae that have moved from obvious feeding sites also could be collected. Weed hosts showing feeding damage were picked or cut off at ground level and shaken into a 20-liter bucket. Dislodged larvae were collected from the bucket. Weed hosts most prone to *L. subjuncta* infestations and thus most sampled were lamb's-quarter, *Chenopodium album* L.; sow thistle, *Sonchus* sp.; bindweed, *Convolvulus arvensis* L.; and mallow, *Malva neglecta* Wallroth. Approximately 25 infested weeds were sampled at each orchard visit. However, this number was influenced by the amount of weeds in the orchard and the relative infestation levels of trees and weeds, which varied from orchard to orchard. Larval sampling was set at 2 person-h per day, and samples were conducted two times per week (4 person-h per week). Larvae were collected and returned to the laboratory where head capsule width and instar stage were recorded (Doerr et al. 2002). We collected 3,552 larvae over the duration of this study.

Temperature data were collected at each site by placing a max-min temperature recorder (Datascribe Jr., Avatel, Inc., Fort Bragg, CA) inside white Stephenson weather shelter. The weather shelter was placed within the canopy of a tree in the interior of the orchard. Degree-day accumulations at each site were calculated using a single sine-wave method (Baskerville and Emin 1969) from daily maximum and minimum temperatures and a base threshold of 6.7°C (Doerr et al. 2002). Degee-day accumulations began at all locations on 1 March. The upper threshold for development and the degree-day accumulation cutoff method were established using male moth flight data. The best combination of upper threshold and cutoff method was determined by minimizing the variability between predicted and observed start of second generation moth flight at each location. A series of upper development thresholds (28, 29, 30, 31, 32, 33, and 34°C) was evaluated using either a horizontal or vertical cutoff method.

Emergence predictions for *L. subjuncta* flight and subsequent egg hatch of each generation were based on the Weibull distribution. The Weibull distribution was chosen for its adaptability and the ease with which

Table 1. Mean absolute deviation in days and SEM predicting second generation flight of *L. subjuncta* (1998–2001) by using different upper thresholds and cutoffs for the sine-wave method of degree-day calculation

	Upper threshold for development (°C)											
	Horizontal cutoff							Vertical cutoff				
	28	29	30	31	32	33	34	30	31	32	33	34
Avg | pred-obs | [a]	2.98	2.65	2.53	2.46	2.39	2.42	2.42	10.88	7.07	4.49	3.09	2.51
SEM	0.29	0.26	0.24	0.22	0.21	0.22	0.21	0.64	0.51	0.41	0.31	0.24

$n = 57$ total orchard-yr.
[a] Mean absolute deviation (predicted minus observed calendar day).

it can be fit to the data set. The Weibull distribution was fit as follows:

$$f(x) = 100 \times \left[1 - \exp\left(-\left(\frac{DD}{\beta}\right)^{\alpha}\right)\right]$$

where $f(x)$ is the cumulative percentage of emergence, DD is the degree-days at which a particular percentage emergence was observed in the field, α affects the shape of the emergence curve, and β is a scale factor. The estimates of α and β were determined by maximum likelihood estimation of the equation by using JMP software (SAS Institute 2002). Previous work with other insects (Evenden and Judd 1999; V.P.J., unpublished data) suggested that the fit of the Weibull distribution lagged behind the early part of the flight curve observed in the field. To reduce this problem, a weighted regression was used where the first 12.5% of the population was weighted 4 times higher than data occurring after that point. This increased the fit at the early part of the flight with virtually no discernible effects later in the generation.

Moth catches from five orchards that maintained high populations through two growing seasons (10 orchard-yr) and received limited insecticide inputs, especially treatments targeting lepidopteran pests, were used to fit the Weibull distribution. First generation egg hatch data taken from six high-density orchards were also fit to the Weibull distribution. For second generation egg hatch, the Weibull distribution was fit based on forward and backward estimations of observable data. First, hatch was estimated from adult flight data allowing for a protandry plus preoviposition period (determined in this study) and 74.6 DD for egg development (Doerr et al. 2002). Second, hatch estimates were based on the relative abundance of first instars collected in larval samples.

The accuracy of the Weibull distribution to model adult emergence was assessed for emergence between 5 and 95% by using indifference bands of ± 7 d (Welch et al. 1981). Indifference bands were set at ±7 d because this is the interval at which traps were monitored. We feel using indifference bands set closer than ± 7 d would result in errors based solely on experimental design rather than variation in individual or population development. Model validation was done between 5 and 95% because of the difficulty of mathematical models to describe the tails of the flight. Model validation was done on a different set of orchards than those used to develop the model. Adult emergence in eight orchards from 2000 to 2001 (10 total orchard-yr) that were completely independent of the data used to generate the model was used for validation. The sites selected for validation were commercially managed apples (multiple cultivars) with varying population levels. To determine the error rate of the adult emergence models the predicted day of an event (cumulative percentage of trap capture) was plotted against the observed day of the event. Any point that fell outside the indifference band was considered a prediction error. The percentage of time errors occurred was used as a measure of the reliability of the model for *L. subjuncta* management. Egg hatch predictions were not validated because the data set was limited and the difficulty of finding large numbers of egg masses made validation at multiple sites problematic.

Results

The average first capture of adult males occurred at 216.2 ± 2.6 (SEM) degree-days from 1 March. The mean absolute deviation of first male capture was 2.7 ± 0.2 d from 216.2 DD. Oviposition (protandry plus preoviposition period) started at an average of 160.0 ± 7.7 DD from the start of adult flight, or 370.7 ± 11.4 DD from 1 March. The mean absolute deviation of the beginning of oviposition was 2.1 ± 0.4 d from 370.7 DD. First generation egg hatch began at 427.0 ± 12.4 DD from 1 March with a mean absolute deviation of 3.0 ± 0.6 d from 427.0 DD.

The protandry plus preoviposition period (160.0 DD) and degree-day requirements for egg-pupa (820.7 DD, Doerr et al. 2002) were used to estimate the time between initial male activity of the first generation and the start of the second generation at 980.7 DD. An upper threshold for development of 32°C by using a horizontal cutoff provided the least variable predictor of the start of the second flight (Table 1). Mean absolute deviation of the predicted and observed calendar day for the start of the second generation flight was 2.39 ± 0.21 d. The average first capture of adult males of the second generation began at 1217.1 ± 8.3 DD from 1 March. The mean absolute deviation of first male captures varied 2.2 ± 0.3 d from 1217.1 DD. These data from 57 orchard-yr showed little variation between sites and years when degree-day accumulations started at 1 March and indicated that standardizing degree-day accumulations at initial capture (i.e., setting degree-days to zero at adult biofix) was unnecessary.

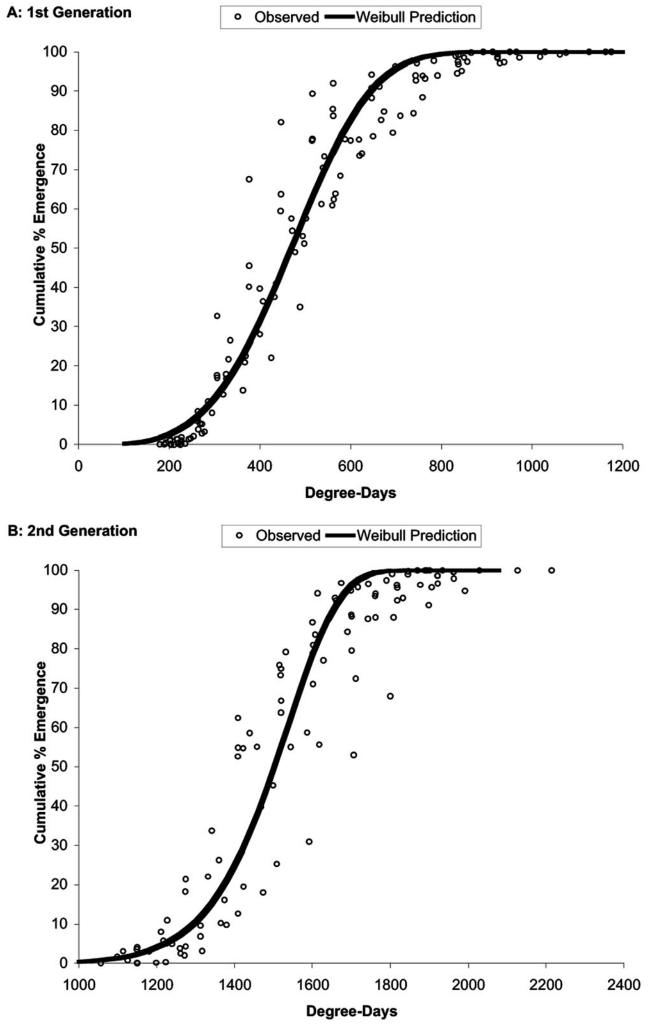

Fig. 1. Cumulative adult emergence during the first (A) and second (B) generations of *L. subjuncta*.

Observed cumulative emergence of *L. subjuncta* adults and cumulative egg hatch were plotted against degree-days (Figs. 1 and 2) along with the Weibull distributions that predicted these life stages. Indifference band validation plots were presented for adult flight only (Fig. 3). First generation adult flight (Fig. 1A) was set to begin at 160 DD (1% predicted emergence), with 5, 50, and 95% emergence predicted at 240, 470, and 700 DD, respectively ($\alpha = 3.8 \pm 0.01, \beta = 516.6 \pm 0.6$). Second generation adult flight (Fig. 1B) was set to begin at 1080 DD, with 5, 50, and 95% emergence predicted at 1220, 1500, and 1690 DD, respectively ($\alpha = 12.5 \pm 0.07$, $\beta = 1546.9 \pm 0.7$). An outlier population was noted during each generation. One population emerged earlier than the others during the first generation and one emerged later during the second generation. With no apparent condition that would explain these variations, they were left in the data set and treated as normal variance between populations. Indifference band validation plots showed a 19.0 and 13.6% error rate for first (Fig. 3A) and second (Fig. 3B) generation adult emergence, respectively. Most error during the first generation was still closely associated with model predictions (just outside the ±7-d bands and evenly distributed both early and late). Second generation errors were clustered toward the end of the flight and generally were the result of flight continuing past the predicted end of the flight. This error is generally regarded as insignificant as the most important portion of the model for pest management decisions is the early part of the flight. First generation egg hatch (Fig. 2A) was set to begin at 330 DD, with 5, 50, and 95% egg hatch predicted at 395, 530 and 630 DD, respectively ($\alpha = 8.8 \pm 0.1$, $\beta = 549.7 \pm 0.6$). The predicted cumulative egg hatch was well correlated to a backward estimation of observed first instar collections. First instars were initially collected at 388 DD, with 5, 50, and 95% of all first instars collected by 475, 500, and 675 DD, respectively (Table 2). Second generation hatch pre-

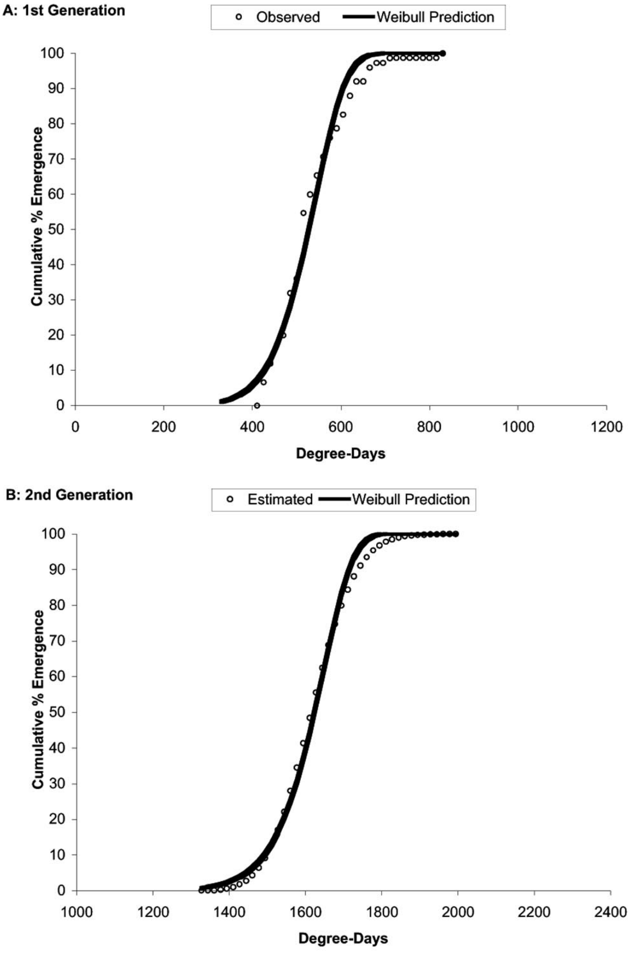

Fig. 2. Cumulative egg hatch during the first (A) and second (B) generations of *L. subjuncta.*

dictions were based on a series of estimations from the observable data associated with adult flight and larval collection. First, we predicted hatch to start at 1451 DD, calculated by adding the 1217.1, 160.0, and 74.6 DD requirements for the observed start of second flight, protandry plus preoviposition, and egg development, respectively. Furthermore, second generation flight was predicted to be 95% complete by 1690 DD, indicating the majority of oviposition would be complete by that time. Next, first instar larvae were initially collected at 1425 DD, with 5, 50, and 95% collected at 1425, 1575, and 1725 DD, respectively (Table 2). Second generation egg hatch modeling (Fig. 2B) was set to begin at 1340 DD, with 5, 50, and 95% hatch predicted at 1440, 1630, and 1740 DD, respectively ($\alpha = 22.2 \pm 0.2$, $\beta = 1650.8 \pm 0.7$). These parameters best placed the majority of estimated egg hatch based on L1 collections and flight activity within a predicted 5–95% cumulative hatch, discounting the expected tails at either end of the Weibull distribution. It should be noted that larvae became relatively more difficult to collect as they aged, illustrated by the fewer number collected of each successively older instar (Table 2). This was likely the result of larvae distributing themselves throughout the tree canopy and orchard ground cover as they matured and eventually entering the soil to pupate during the sixth instar. It is unlikely that this fact changes observations about population development but rather makes relative comparisons between instar stages difficult as all collections are biased toward younger larvae.

Discussion

The sudden rise in pest status of *L. subjuncta* in central Washington and northeastern Oregon apple orchards forced growers, crop consultants, and researchers to quickly develop a pest management approach to integrate into the existing program. Laboratory bioassays and field trials can indicate insecticides that are efficacious, but the most difficult decisions faced by growers and crop consultants are determining what qualifies as a treatable population and when to apply control treatments. With little detailed information on a pest's phenology, sampling for larvae and timing insecticide applications was rel-

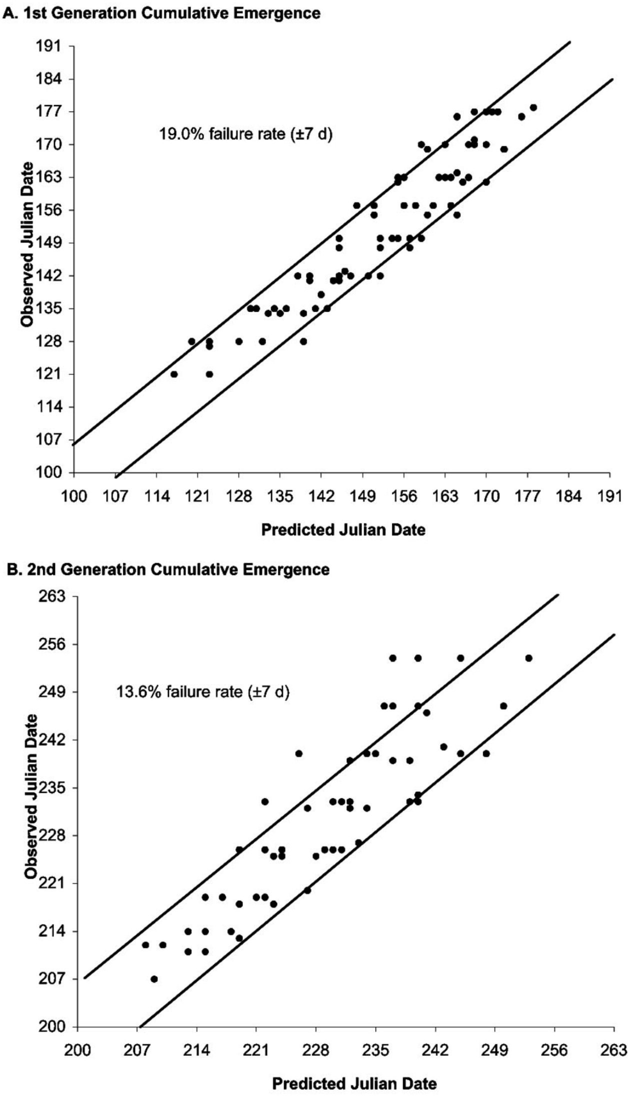

Fig. 3. Indifference band validation plots of the cumulative emergence (5–95%) of first (A) and second (B) generation *L. subjuncta* adults using indifference bands of ±7 d.

egated to a trial-and-error exercise. The result has often been an increase in the rates and number of times pesticides are applied, which increase costs for the grower and potentially negatively impact biological control activity in the system.

Degree-day models have been used successfully in tree fruit production for many years (Welch et al. 1978, Whalon and Croft 1984). These models are especially useful for predicting the occurrence of an event in a pest's life cycle that is difficult to monitor. The detailed phenology descriptions presented here, especially egg hatch and larval age distributions, are expected to make the timing of insecticide applications more accurate and therefore more effective. For example, the ideal timing for the application of a "soft" pesticide with relatively short residual activity, such as spinosad (Success 2 SC, Dow AgroSciences LLC, Indianapolis, IN), would be when the majority of egg hatch was complete and most larvae were of a susceptible stage, third instar or earlier (Doerr et al. 2004). First generation egg hatch was estimated to be 80% complete by ≈580 DD (Fig. 2A). At 650 DD, all larvae collected in field samples were in the first through third instars (Table 2). The presence of more mature larvae, fourth through sixth instars, was not noted until 675 DD. These data indicate that the best timing for an insecticide application would be 575–650 DD. A 75 DD window at this time of year equates to an average of 10 calendar days based on 30-yr average temperatures in central Washington (Washington State University Tree Fruit Research and Extension Center, Wenatchee, WA). This period provides sufficient time to apply an insecticide under acceptable climatic conditions. During the second generation,

Table 2. Cumulative degree-days in 25-unit increments from 1 March and associated *L. subjuncta* larval collections during the first and second generations, 2000–2001

Generation	n	Accumulated % of total collections for each larval instar							
		1%	5%	10%	25%	50%	75%	90%	95%
First									
1	1,780	425	475[a]	475[a]	500	500	550	600	675
2	231	525	525	550	625	675	750	875	975
3	213	575	600	600	675	875	975	1050	1075
4	209	675	700	750	875	925	1075	1125	1250
5	128	675	750	875	925	1000	1125	1250	1375
6	53	700	700	700	1000	1125	1125	1250	1500
Second									
1	650	1425	1425	1425	1450	1575	1575	1725	1725
2	81	1550	1625	1625	1650	1650	1725	1750	2025
3	68	1625	1625	1650	1725	1750	2025	2125	2150
4	55	1650	1725	1725	1900	2000	2125	2150	2200
5	46	1775	1900	1900	1975	2075	2150	2250	2250
6	38	1900	1900	1900	1975	2000	2250	2300	2300

[a] Multiple instar accumulations with the same degree-day interval were a result of an accumulated percentage for more than one category being reached in the same interval.

80% egg hatch was estimated at 1690 DD (Fig. 2B). At 1700 DD, all larvae collected in field samples were still first through third instars, with the exception of one fourth instar (Table 2). The presence of fifth and sixth instars was not noted until 1775 DD. These data indicate that the best timing for an insecticide application against larvae of the second generation would be 1650–1725 DD. This represents a period of seven calendar days based on 30-yr-average temperatures in central Washington.

Another use of the model is to provide growers and crop consultants with a tool to identify a sampling period just before the predicted time for optimum insecticide applications for assessing larval densities or feeding activity on which control decisions can be made. By sampling just before the windows discussed above and estimating the number of larvae or feeding damage the need for control decisions can be made and the timing of effective insecticides optimized to achieve economic control of *L. subjuncta* with only one application per generation (Doerr et al. 2004).

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