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**HEAD CAPSULE WIDTHS AS AN INDICATOR OF THE LARVAL INSTAR OF CODLING MOTH (LEPIDOPTERA: OLETHREUTIDAE)<sup>1</sup>**Peter Weitzner<sup>2</sup> and Mark E. Whalon<sup>3</sup>**ABSTRACT**

Head capsule width was a reliable indicator of larval instar in a strain of Michigan codling moths, *Cydia pomonella*. Head capsules were 0.33, 0.50, 0.82, 1.18 and 1.55 mm in width from first to fifth instar respectively. Development as measured by days and degree days was much more variable than head capsule width in estimating larval instars.

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One of the most important insect pests of apples in North America is the codling moth, *Cydia pomonella* (L.). In late spring the overwintering, late instar larvae come out of diapause, resume development, and pupate. Within 20–25 days, at optimum conditions, first generation adults emerge and lay eggs. Eggs are laid on or near the fruits, and newly hatched larvae crawl across the surface of the fruit to locate an entry site. After entry sites are located, the larvae bore directly into the fruit toward the seeds and feed there until their larval development is complete. The late instar larvae (fourth or fifth) emerge from the fruit and either seek an overwintering site where they spin a hybernaculum and overwinter, or continue development through the pupal stage and emerge as adults to complete the cycle.

In Michigan apple orchards the codling moth causes an average of 0.52% damage to commercial apples (CCMS 1984) in spite of sophisticated phenology models (Welch et al. 1978, Riedl and Croft 1978, Gage et al. 1982) which are used to time insecticide sprays. A research program was initiated in 1982 to improve the predictive capability of this phenology modeling system by including the effects of temperature modification on diapause induction (Garcia-Salazar et al., in press). Several of these studies and subsequent field validation of the models required a means of estimating instar without knowing the developmental history of the larva. There are no reliable morphological traits for distinguishing different codling moth larval instars except size and the sexual dimorphism apparent only in the fifth instar (Hansen and Harwood 1968). Larval head capsule width in other Lepidoptera (eg. *Choristoneura rosaceana* (Harris) and *Argyrotaenia velutinana* (Walker)) has been a reliable means of estimating developmental instars. Since these data were not available for the Michigan strain of codling moth, our objective of this study was to correlate head capsule width with larval instar.

**MATERIALS AND METHODS**

Michigan-strain adult codling moths originating from Douglas, Michigan, in 1982 were used in this study. Adults from this regularly maintained culture (Garcia-Salazar 1984)

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were placed into circular plastic (21 × 9-cm) oviposition containers. The containers were lined with wax paper as an oviposition surface. Adult moths were fed 50% sucrose solution contained in a 10-ml specimen vial equipped with a cotton dental wick. The codling moth colony and experimental larvae were held in a growth chamber at 27°C, 70% RH, and 16L:8D photoperiod. A twilight was produced before and after each light photoperiod by a small microscope light (200 lux) which remained on for an additional 20 min.

A semisynthetic diet (Cowles 1985) was prepared and poured into individual 1.25-oz cups. Both the cups and their covers were sprayed with 70% ethanol solution with methyl paraben (2%) and solidified under an ultraviolet light. When dry, the surface was scratched with a sterile knife and a first instar larva was transferred to the media with a small, soft-bristled brush. Pieces of cardboard (1.5 × 1.5 cm) were glued to the covers of the cups to provide the larvae with a pupation site.

Treatment larvae were selected by randomly identifying a cohort of eggs laid within 1 h of each other and examining them at 1-h intervals for hatching. When hatching occurred, the larvae were placed into the diet cups immediately. Each larva's cup was dated and the time recorded at egg hatch.

Forty Larvae were randomly selected and assigned to each of eight treatments of 2, 4, 6, 8, 10, 12, 14, and 16 days of development time (total 320 larvae). At the end of each period, the treatment cups were opened and the head capsules of the larvae were measured and recorded. A dissecting binocular microscope with a micrometer ocular was used. Data were submitted to a one-way analysis of variance (SAS 1985) and mean separation procedures (Steel and Torrie 1982).

Examination of the data suggested fitting cumulative density functions to each instar's heat unit requirements for completion of a developmental stage. Cumulative density functions predicted the fraction of the total larvae in a particular instar predicted by the cumulative density functions in each treatment group. The density functions were used to estimate the developmental time (days) and degree day (DD) summations.

## RESULTS AND DISCUSSION

Head capsule measurements made differentiation between instars very clear. Molting resulted in a significant ( $P \leq 0.01$ ) increase in head capsule width (Table 1) as expected, allowing for clear distinction between instars. Head capsule widths within instars varied over a small range as demonstrated by the small standard deviations. The variability in head capsule width data shows that 75.2 and 95.4% of the values fall within one and two standard deviations of the mean respectively.

Since the requirement of sterile conditions in this experiment allowed only one measurement to be made on a larva, the determination of development time and DD requirements (continuous variables) for a particular larva were not possible. Therefore, the most effective alternative method of determining these parameters was to develop an analysis of the cumulative frequency functions of each instar on the respective observation days. We reasoned that average development time would occur at or around 50% completion of a developmental stage, therefore, determination of mean development time and DD were used to estimate 40 data points for each instar. Mean developmental time and DD were calculated from these data. Developmental time (days) and DD per stage were much more variable. Each successive instar required greater thermal units to complete development, except the fifth instar, the shortest developmental stage, which required only  $35.2 \pm 61.83$  DD to complete. Developmental time in each larval instar also progressed from first to fourth instars, and the fifth instar again required the shortest time (1.1 days). Total instar developmental time averaged 14.4 days.

Figure 1 presents the cumulative density functions for each instar. Since the larvae were all in the first instar when the experiment started, these data are presented as a decreasing density function while second through fifth are all increasing. The measurement of first instar development is underestimated since all of the larvae were first instar at 0 DD (i.e.,

Table 1. Mean ( $\pm$ SD) of head capsule widths (mm), development time in days and degree days ( $\pm$ SD) base 50°F for the five larval instars of the Michigan strain of codling moth.

Larva	Head Capsule Width (mm) <sup>a</sup>		Time in Stage (Days)		Degree Days	
	$\bar{x}$	( $\pm$ SD)	$\bar{x}$	Range	$\bar{x}$	( $\pm$ SD)
1st Instar	0.33 <sup>A</sup>	0.017	2.7	1.4-4.0	86.9	41.81
2nd Instar	0.50 <sup>B</sup>	0.027	3.1	1.1-4.9	98.0	61.50
3rd Instar	0.82 <sup>C</sup>	0.063	3.2	0.7-5.8	103.8	82.56
4th Instar	1.18 <sup>D</sup>	0.072	4.3	2.2-6.5	138.6	68.82
5th Instar	1.55 <sup>E</sup>	0.073	1.1	0.6-3.0	35.2	61.83

<sup>a</sup>Means followed by the same letter are not significantly different ( $P < 0.01$ ), LSD test (Steel and Torrie 1982).

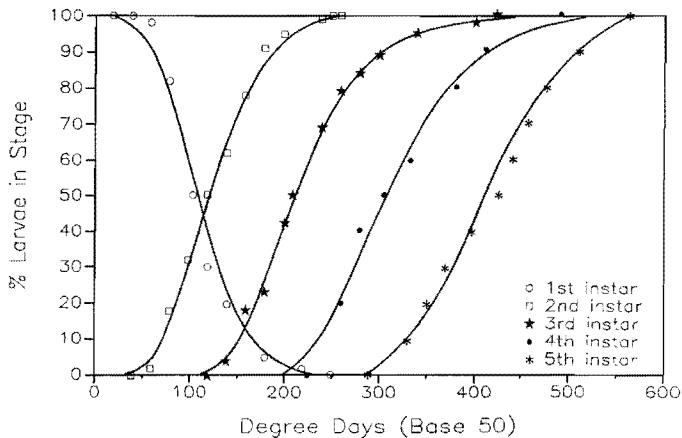


Fig. 1. The cumulative percent of larvae found in each developmental stage (1st-5th larval instars) at various days and degree days (base 50°F).

initiation of the experiment). It took an average of 462.5 DD to complete development from first to the fifth instar larvae. This compares well with the 510 DD (Base 50°F) reported by Rock and Shaffer (1983). These developmental estimates are similar given the different experimental designs and the genetic variability in developmental time observed in other codling moth strains (Garcia-Salazar 1984). The average cumulative development data were used in the development of a model to predict the influence of temperature and photoperiod for diapause induction in the codling moth (Garcia-Salazar et al., in press).

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