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AN ALLOMETRIC STUDY OF THE BOXELDER BUG, *BOISEA TRIVITTATA*  
(HETEROPTERA: RHOPALIDAE)Scott M. Bouldrey and Karin A. Grimnes<sup>1</sup>

## ABSTRACT

An allometric study was conducted on the boxelder bug, *Boisea trivittata*, to confirm the number of instars and to identify characteristics most useful for rapid instar identification of field samples. Analysis of field populations collected throughout the 1990-92 seasons indicated that there were five instars, clearly defined on the basis of size and the presence of wing pads. This finding is in contrast with the only other published study on stages of the boxelder bug, which claims there are six nymphal instars.

Size data gathered from field populations were substantiated by laboratory growth studies. Head width and/or second antennal segment length were identified as the most useful parameters for instar identification.

*Boisea trivittata* (Say), the boxelder bug, is a pest closely associated with the pistillate boxelder tree (*Acer negundo*) throughout much of their shared range (Slater and Schaefer 1963). During hot, dry summers, the number of boxelder bugs can reach epidemic proportions, as evidenced by a long history of scattered reports in the literature (see the review in Wheeler 1982). Although they rarely cause economic damage, boxelder bugs are considered a significant household nuisance.

A recurring outbreak site in Gratiot County, Michigan, was discovered in the spring of 1990. As we began population studies, we realized that the only detailed report on this insect's life history identifies six nymphal instars after hatching (Smith and Shepherd 1937). According to these authors, minute wing pads did not appear until the fourth instar, and the authors did not identify any exuvia left in the egg after hatching (an egg-bound molt). The general rule for hemipteran species appears to be five nymphal instars before the adult ecdysis, not counting any egg-bound molt left after hatching (Slater and Baranowski 1978). Subsequent investigators of this insect either avoided a direct challenge of the instar number (Tinker 1952) or divided the population into only adults and nymphs (Yoder and Robinson 1990). We became interested in the question of instar number for the boxelder bug because our preliminary studies of field populations showed only five clearly discernable instars that were not easily reconciled with results of Smith and Shepherd (1937). As a result, we began a detailed growth study and allometric analysis of our population of *B. trivittata* for use in subsequent seasonal studies.

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### MATERIALS AND METHODS

Insects were collected at Lang's Veterinary Clinic west of Alma in Gratiot County, Michigan, biweekly from March emergence to October hibernation during 1990-1992. The site contains numerous staminate and pistillate box-elder trees (*Acer negundo*) close to the residence. Insects were swept into 70% ethanol with a small brush from the same stretch of south-facing wall and west-facing fence on each collecting date.

Samples were separated into five nymphal instars or adults on the basis of body size, presence and degree of wing pad growth, and head width. All measurements were made under a dissecting microscope with an ocular micrometer. Body parts (see Tables 1 and 2) were measured at their maximum point with the exception of antennal segments (1-4) where only the melanized portions were measured. For each instar, and for adult males and females, thirty-five individuals were measured.

Mated females were allowed to oviposit in the laboratory on filter paper in 100 mm petri dishes. Because mortality of individually raised animals was high, first instars were kept in groups of five in petri dishes containing moistened filter paper. Maple syrup (5% v/v) was loaded into cotton-plugged tubing and placed in the dish for food. The dishes were exposed to a 16:8 LD photoperiod and maintained at  $24^{\circ}\text{C} \pm 1^{\circ}\text{C}$  in an incubator. Animals were observed daily, molts were recorded, and the newly molted animals were moved to new dishes (in groups of five if possible). Head widths and second antennal segment lengths were recorded for recently molted individuals.

Means between morphological characteristics of male and female adults, and between field and laboratory-raised animals, were compared with student's t-test. Growth curves were fit using linear regression techniques.

### RESULTS

Size of nymphs is highly correlated with age for all parameters measured in this study (Table 1). When growth data for the five instars were analyzed with simple linear regression, no parameter had an  $r^2$  value less than 0.90. Head width was most highly correlated to instar number, as was length of the second antennal segment. For pronotal length and width, visual inspection of the data indicated that a logarithmic curve might better describe the growth relationship. Subsequent analysis of the logarithmic data produced an  $r^2$  value of 0.96 (pronotum length) and 0.97 (pronotum width). All correlations reported in this study are somewhat time independent because the regression analysis is against instar number (which assumes equal duration of each stage) and not against actual growth in days.

Adult females were significantly larger than adult males for all parameters measured (Table 2). Difference in body length was easily discernable in the field. No attempt was made to sex fifth instars in this preliminary study, so some of the variation in fifth instar measurements could result from sex-related growth differences.

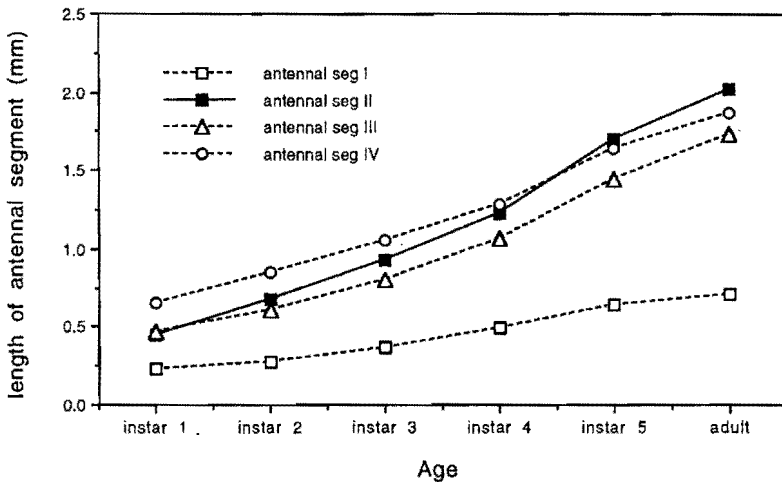
Although head width classically is used as an indicator of growth for many insect species, we noticed that the length of the second antennal segment was equally predictive of instar number in *B. trivittata* (Table 1). This observation led us to compare growth rates (using instar number, and not actual time) of the four antennal segments (Fig 1). Clearly, the first segment changes little as the animal grows, but all other segments lengthen noticeably with instar. The second segment is the second shortest at hatching, yet by the fifth instar,

Table 1. Measurements of nymphs of *Boisea trivittata* (Say). Values are mean mm  $\pm$  SE, n=35 (based on field collected individuals).

Character	1st Instar	2nd Instar	3rd Instar	4th Instar	5th Instar	r <sup>2</sup>	slope
Body length	2.91 $\pm$ 0.04	3.80 $\pm$ 0.06	5.11 $\pm$ 0.06	7.01 $\pm$ 0.09	9.60 $\pm$ 0.11	0.93	1.67
Head width	0.68 $\pm$ 0.01	0.92 $\pm$ 0.01	1.17 $\pm$ 0.01	1.51 $\pm$ 0.01	1.89 $\pm$ 0.02	0.97	0.30
Interocular dist.	0.50 $\pm$ 0.01	0.63 $\pm$ 0.01	0.80 $\pm$ 0.01	1.04 $\pm$ 0.01	1.28 $\pm$ 0.01	0.96	0.20
Pronotal length	0.29 $\pm$ 0.01	0.38 $\pm$ 0.01	0.57 $\pm$ 0.01	0.76 $\pm$ 0.01	1.51 $\pm$ 0.01	0.91	0.21
Pronotal width	0.80 $\pm$ 0.01	1.01 $\pm$ 0.01	1.30 $\pm$ 0.01	1.69 $\pm$ 0.02	2.51 $\pm$ 0.03	0.90	0.41
Antennal length							
Segment 1	0.23 $\pm$ 0.01	0.28 $\pm$ 0.01	0.37 $\pm$ 0.01	0.49 $\pm$ 0.01	0.64 $\pm$ 0.01	0.93	0.10
Segment 2	0.45 $\pm$ 0.01	0.68 $\pm$ 0.01	0.93 $\pm$ 0.01	1.23 $\pm$ 0.01	1.70 $\pm$ 0.01	0.97	0.31
Segment 3	0.46 $\pm$ 0.01	0.61 $\pm$ 0.01	0.81 $\pm$ 0.01	1.07 $\pm$ 0.01	1.44 $\pm$ 0.01	0.95	0.24
Segment 4	0.65 $\pm$ 0.01	0.85 $\pm$ 0.01	1.05 $\pm$ 0.01	1.28 $\pm$ 0.01	1.64 $\pm$ 0.01	0.95	0.24

Table 2. Measurements of adults of *Boisea trivittata*. Values are mean mm  $\pm$  SE, n=35 (field collected individuals).

Character	Adult females	Adult males	p values <sup>a</sup>
Body length	12.1 $\pm$ 0.10	10.3 $\pm$ 0.08	<0.0001
Head width	2.14 $\pm$ 0.01	1.96 $\pm$ 0.02	<0.0001
Interocular dis.	1.34 $\pm$ 0.01	1.19 $\pm$ 0.02	<0.0001
Pronotal length	1.93 $\pm$ 0.04	1.73 $\pm$ 0.02	<0.0001
Pronotal width	3.70 $\pm$ 0.03	3.09 $\pm$ 0.02	<0.0001
Antennal length			
Segment 1	0.64 $\pm$ 0.01	0.62 $\pm$ 0.01	<0.01
Segment 2	2.09 $\pm$ 0.02	1.90 $\pm$ 0.01	<0.0001
Segment 3	1.80 $\pm$ 0.02	1.70 $\pm$ 0.02	<0.0001
Segment 4	1.90 $\pm$ 0.02	1.73 $\pm$ 0.02	<0.0001

<sup>a</sup> student's t-test.Figure 1. Mean length of antennal segments (mean  $\pm$  SE) of field-collected *Boisea trivittata* specimens. Thirty-five measurements were averaged for each nymphal data point; values for adults were obtained by averaging male and female means from Table 2.

it is the longest. In the regression analysis, the predictive growth line for the second antennal segment has the greatest slope (Table 1, Fig. 1).

Growth studies in the laboratory were carried out on animals reared in groups of five, wherever possible. Animals reared as individuals perished quickly; only eight of 58 (13.8%) molted to the second instar, and none of those animals survived to the third instar. In contrast, 43.1% (148/343) of animals

raised in groups of four or five survived to the second instar, and 17.0% (8/47) survived to the third instar. Mass rearing (even in groups of as few as five) increases the risk of death from cannibalism, a phenomenon reported first by Abbott (1948). In our study, cannibalism appeared to be rare. It was observed only in the absence of a water source and only when an animal emerged unusually helpless and unable to move away from the probosci of its fellow hatchlings.

We found no evidence of egg-bound exuvia after first instars hatched from the egg. Under our laboratory conditions, the first stadium averaged  $5.5 \pm 0.2$  days ( $n=148$ ) and first instars had a mean head width of  $0.64 \pm 0.01$  mm ( $n=25$ ) and a mean second antennal segment length of  $0.42 \pm 0.01$  mm ( $n=25$ ). The second instars had a mean head width of  $0.88 \pm 0.01$  mm ( $n=20$ ) and a mean second antennal segment length of  $0.66 \pm 0.01$  mm ( $n=20$ ), while the mean instar duration was  $6.5 \pm 0.8$  days ( $n=8$ ). For the third instars, the mean head width was  $1.09 \pm 0.01$  mm ( $n=6$ ), while the mean length of the second antennal segment was  $0.92 \pm 0.01$  mm ( $n=6$ ). All third instar nymphs possessed wing pads, in contrast to the study of Smith and Shepherd (1937) where wing pads were not observed until the fourth instar. Although the mean values of all parameters measured were slightly smaller for insects raised in the laboratory, neither head widths nor second antennal segment lengths were significantly different from field-caught instars (student's t-test). Unfortunately, all laboratory-reared animals were lost because of an incubator malfunction just before the majority of insects reached the third instar, resulting in a low number of data points ( $n=6$ ) for measurements of that instar. Nevertheless, the presence of wing pads in lab-reared third instars clearly was established.

## DISCUSSION

There has not been a detailed growth analysis of the boxelder bug previous to the present study. Smith and Shepherd (1937) reported head width and body length data, but included only mean and range values. Also, as previously mentioned, they identified six, rather than five, instars for this insect. We believe we have clearly established the presence of five, and only five, instars in *B. trivittata*, not only through our identification of discrete size classes for the parameters measured in field animals, but through the insects that were reared into the third instar. Although Smith and Shepherd report duration data for six instars, their sample numbers suggest that the same individuals were not followed throughout their entire life cycle.

All of the third instars raised in our study possessed wing pads (often considered "the rule" for hemipteran species) gives the first clue toward explaining the differences between our data and those of Smith and Shepherd (1937). Comparing mean head widths for our third instar (1.17 mm) to their fourth (1.17 mm), our fourth instar (1.51 mm) to their fifth (1.51 mm), and our fifth instar (1.89 mm) to their sixth (1.83 mm) makes it clear that the discrepancy in instar identification must lie prior to our third instar. Also, mean head width for our second instar sample (0.92 mm) matches their third instar (0.90 mm), and if we average their values for first (0.64 mm) and second instars (0.78 mm), the head width of 0.71 mm compares nicely with our first instar value of 0.68 mm. No statistical differences were seen between our field and laboratory-reared populations for either head width or for length of the second antennal segment for instars 1 - 3, further substantiating our proposed instar identification of field-caught animals. Based on these data, we believe that Smith and Shepherd may have inadvertently subdivided a highly variable first instar population into smaller than average (instar 1) and larger

than average (instar 2) samples and labelled them accordingly. We observed that slightly desiccated eggs may result in hatchlings that do not expand before sclerotization to the same extent as nondesiccated animals; thus, desiccation results in animals with smaller head widths and a wider variation in hatchling size.

The values we report for adult head width, pronotum length and width, and length of the four antennal segments agree fairly well with the values presented by Schaefer (1975) for six populations of *B. trivittata* collected across the United States. His samples of adults were not identified by sex: slight differences in mean values, therefore, may represent variation in the sex ratios of his samples. Both male and female adults observed by Smith and Shepherd (1937) possessed slightly larger dimensions for head width, body length, and pronotal width than the adults in this study.

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