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# TEMPORAL DISTRIBUTION OF EGG HATCH FOR TWO *HOMALODISCA* SPP. (HEMIPTERA: CICADELLIDAE) UNDER CONSTANT TEMPERATURES

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The glassy-winged sharpshooter (GWSS), *Homalodisca vitripennis* (Germar) (=*H. coagulata* (Say)) (Takiya et al. 2006), is an exotic pest in California and an important vector of the xylem-limited bacteria, *Xylella fastidiosa* Wells et al., the causal agent of several plant diseases including Pierce's disease of grapes, phony peach disease, almond leaf scorch, alfalfa dwarf, and oleander leaf scorch (Blua et al. 1999; UCOP 2000; Varela et al. 2001). The closely related smoke-tree sharpshooter (STSS), *H. liturata* Ball (=*H. lacerta* (Fowler)) (Burks & Redak 2003), is native to California and also a vector of Pierce's disease and oleander leaf scorch (Freitag et al. 1952; Purcell et al. 1999). Eggs of both *Homalodisca* spp. are laid just below the epidermis of leaves and other plant parts including stems and fruits in clusters, with eggs oriented nearly parallel to one another.

Al-Wahaibi & Morse (2003, 2009) described the relationship between the rate of embryonic development of *H. vitripennis* and temperature in the range of 16.7-35.0°C. We present here a follow-up study aimed to elucidate the distribution of emergence of first instars of *H. vitripennis* and *H. liturata* across time, and to determine whether different constant egg incubation temperatures would modify that distribution. In order to satisfy these objectives, 2 experiments were conducted. The first examined the daily distribution of egg hatch of *H. vitripennis* and *H. liturata* at 8 constant temperatures. The second evaluated the diurnal distribution of hatching of *H. vitripennis* at 3 constant temperatures.

For the first experiment, egg masses were collected by caging field-collected adults of *H. vitripennis* on potted, rooted cuttings of chrysanthemum, *Dendranthema grandiflorum* (Kitam) (White Diamond cultivar, Growlink Co., Ventura, CA), in sleeve cages which were held at 23°C, 50-70% RH, and 14:10 L:D photoperiod inside an insectary room. Separate plants were exposed to colonies of the 2 leafhopper species for 24 h. Plants with egg masses of the 2 species were then incubated at the same time *in situ* (inside leaves intact on plant) in growth chambers (Percival Scientific, Inc., Perry, IA) at 13.0, 16.7, 19.7, 25.6, 31.2, 32.9, 33.4, and 35.0°C (mean temperatures based on HOBO data loggers, Onset Computer Co., Bourne, MA). For all temperature treatments, relative humidity varied between 50-70%, and the light regime was set at 14:10 L:D photoperiod.

When hatching was imminent (presence of large dark eye-spots), leaves containing egg masses were excised and placed inside 100-mm diameter Petri dishes on top of moist tissue paper. This allowed easier and more accurate observation of emergence of nymphs from individual eggs. Egg masses inside Petri dishes were retained at the same incubation temperatures as the egg masses were exposed to prior to excision of leaves. Thereafter, egg hatch was monitored twice daily, in the morning (8-10 AM) and afternoon (4-6 PM). The proportion of eggs hatching during the first day of hatch and during the following 6 d (i.e., 7 d in total) was calculated (out of the total number of hatched eggs per egg mass). Each egg mass was used as a replicate. Following this, the proportion of eggs hatching during each of d 1, 2, 3, and 4-7 were compared among temperature treatments by ANOVA. Means shown to be significantly different by ANOVA were further compared to each other and separated with the Tukey-Kramer HSD procedure. In addition, the proportion of eggs hatching (pooled across temperature treatments) during each of d1, 2, 3, and 4-7 were compared between the 2 *Homalodisca* spp. by *t*-tests. All statistical tests were performed with JMP IN (SAS Institute 1996).

Most hatching occurred during the first day (79%). The percentage of eggs hatching during the second day averaged 19%, with 2% on the third day. Combined egg hatch from d 4 to 7 eggs averaged 0.2%. There was no significant difference between the 2 *Homalodisca* species for the proportion of hatched eggs on each of d 1, 2, 3, or 4-7 (all *t*-tests, $P > 0.05$). Temperature had a slightly significant effect on first day hatching (ANOVA, $P = 0.0247$) (Fig. 1). Generally, greater first day hatch was observed at higher temperatures, with hatching at 33.4°C being significantly higher than at 19.7°C. Temperature had a marginally significant effect on the proportion of second day-egg hatch ($P = 0.0502$), with slightly higher hatching at 16.7 and 19.7°C than at the higher temperatures. Third day hatching was significantly higher at 13.0°C than at all other temperatures ($P = 0.0003$). There was no significant effect of temperature on egg hatch over d 4-7 ($P = 0.1553$).

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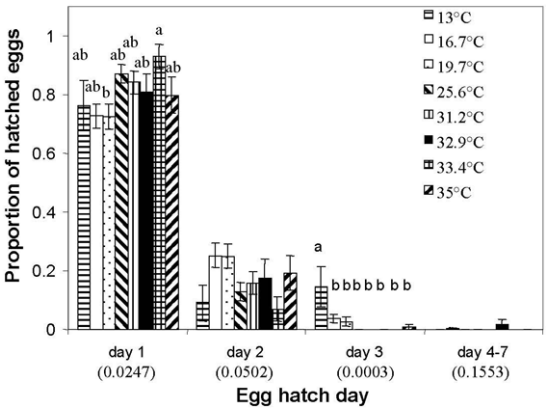



Fig. 1. Effect of temperature (pooled across the 2 *Homalodisca* species) on the proportionate egg hatch for each of the first 3 d of egg hatch and for combined hatch during d 4 to 7. ANOVA derived *P* values are shown below each egg hatching day category. Means for columns indicated with the same letter are not significantly different. Error bars are standard errors of the mean; 13°C, *n*=11; 16.7°C, *n* = 53; 19.7°C, *n* = 62; 25.6°C, *n* = 33; 31.2°C, *n* = 38; 32.9°C, *n* = 20; 33.4°C, *n* = 25; 35°C, *n* = 23.

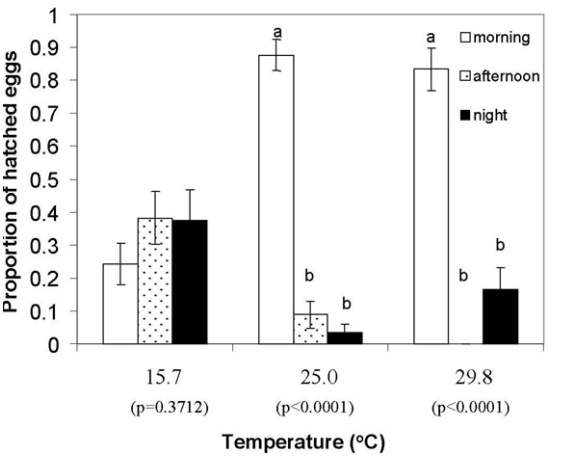



Fig. 2. Diurnal distribution of egg hatch of *H. vitripennis* among 3 daily periods (see text) at 3 different temperatures. ANOVA derived *P* values are shown below each temperature treatment. Columns with the same letter are not significantly different. Error bars are standard errors of the mean; 15.7°C, *n* = 21; 25.0°C, *n* = 40; 29.8°C, *n* = 26.

A second experiment evaluated the effect of temperature on the diurnal distribution of *H. vitripennis* egg hatch. Egg masses were produced by caging field-collected *H. vitripennis* adults on potted seedlings of jojoba (*Simmondsia chinensis* (Link) Schneider) in sleeve cages which were held as before in an insectary room exposed to leafhoppers for 24 h. Plants were then incubated in growth chambers at 15.7, 25.0, or 29.8°C with relative humidity set at 60% and lighting at 12:12 L:D. When hatching was imminent, leaves containing egg masses were excised and placed in Petri dishes as described above. Egg masses were checked thereafter at 6 AM, 12 noon, and 6 PM daily from the day of initial emergence and for the following 3-5 d until no further emergence was observed. For the purpose of analysis, the day was divided into 3-egg hatch periods: a morning period (6 AM to 12 noon), an afternoon period (12 to 6 PM), and a night period (6 PM to 6 AM). Records were kept of the number of hatched eggs (emerged and emerging nymphs) for each of the 3 periods. These data were used to calculate proportionate egg hatch during each of the 3 periods out of the total number of hatched eggs per egg mass; these data were compared across temperatures by ANOVA. Means with significant *F* values were further compared by the Tukey-Kramer HSD procedure.

At 15.7°C, egg hatch did not differ significantly among the 3 daily periods ($P = 0.3712$; Fig. 2). This is in contrast to the pattern for 25.0 and 29.8°C in which there was significantly higher egg hatch in the morning than in the afternoon and night ($P < 0.0001$ for both temperature treatments). For 25.0 and 29.8°C, the afternoon and night levels of egg hatch were not significantly different from one another. However, for the 25.0°C treatment, egg hatch was slightly lower at night than in the afternoon, while at 29.8°C, the night levels of hatch was considerably higher than the afternoon level.

Hoffman et al. (1990) observed that all or most eggs of the potato leafhopper, *Empoasca fabae* (Harris), hatched in the morning. In other leafhopper and insect species, the diurnal rhythm of egg hatch likely depends on temperature, with lower temperatures causing a more equal distribution of emergence over time. Although the hatching process of leafhopper eggs might commence with the first light stimulus at dawn for all temperatures, the hatching of first instars could continue much later in the day when temperatures are cool enough to slow hatching activity. Thus, it appears likely that the temperature at which Hoffman et al. (1990) observed hatching was relatively high (20-30°C), similar to temperatures in the current study (25.0 and 29.8°C) which resulted in a mostly morning egg hatch for *H. vitripennis*.

We hypothesize from above laboratory data that in late winter and early spring, when temperatures are relatively low in the mornings, hatching of *Homalodisca* eggs will be staggered, whereas during late spring and summer when morning temperatures are relatively high, hatching and emergence of first instars will be synchronized, occurring on the same day for the most part. The unhardened cuticle of newly emerged first instars

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could make them more vulnerable to chemical or entomopathogenic control. Control of *Homalodisca* spp. (especially the more pestiferous *H. vitripennis*) could be accomplished more effectively during the morning period when a relatively large proportion of vulnerable first instars have recently emerged. Degree-day models that help predict the time of initial egg hatch of *Homalodisca* eggs (Al-Wahaibi & Morse 2003, 2009) can be combined with knowledge of the temporal distribution of egg hatch to better time chemical or biological control measures against first instars of these pest insects.

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## Summary

The temporal distribution of egg hatch for *Homalodisca vitripennis* (Germar) and *H. liturata* Ball was investigated at different constant incubation temperatures. The 2 *Homalodisca* species did not differ significantly in their daily hatching patterns. Practical implications of these findings for the control of *Homalodisca* spp., especially the pestiferous *H. vitripennis,* are discussed.

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