

Development and survival of *Aulacorthum solani*, *Macrosiphum euphorbiae* and *Uroleucon ambrosiae* at six temperatures

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Abstract

Temperature is one of the most important factors in determining the survival and developmental rate of aphids. The aim of this study was to evaluate the developmental time and survival of the aphid species *Aulacorthum solani* (Kaltenbach), *Macrosiphum euphorbiae* (Thomas) and *Uroleucon ambrosiae* (Thomas) (Rhynchota Aphididae) at six constant temperatures. Tests were conducted on lettuce plants (*Lactuca sativa* L.) in climate chambers at 16, 19, 22, 25, 28 and 31 ± 1 °C, 70 ± 10% RH and 12 h photophase. At high temperatures these species showed lower rates of survival than at low temperatures, and at 31 °C no aphids reached adulthood. The highest survival rates and the shortest developmental times were observed between 16 and 22 °C. The lower temperature threshold (T_b) and thermal constant (K) for *A. solani* and *M. euphorbiae* were 1.09 and 1.05 °C and 142.9 and 144.9 degrees-day (DD), respectively, while for *U. ambrosiae* these parameters could not be estimated. The highest survival rate and short developmental time found at 22 °C indicate that this temperature is optimal for development of *A. solani* and *M. euphorbiae*, whereas the optimal temperature for *U. ambrosiae* is 19 °C.

Key words: potato aphid, foxglove aphid, greenhouse, lower temperature threshold, thermal constant.

Introduction

Lettuce, *Lactuca sativa* L., is one of the most consumed vegetables in Brazil (Luz, 2008), and using the right temperature regime for its production is essential. Low temperatures favor its growth (Filgueira, 2000), but high temperatures negatively affect production in summer in almost all lettuce producing regions of Brazil (Goto, 1997).

Next to temperature, pests, and in particular aphids, are also important factors limiting the production volume and quality of lettuce. In Brazil, lettuce is mainly produced in plastic tunnels, either in the soil or hydroponically, and on average 10 production cycles are realized per year (Bueno, 2005). *Macrosiphum euphorbiae* (Thomas), *Aulacorthum solani* (Kaltenbach) and *Uroleucon ambrosiae* (Thomas) (all three Rhynchota Aphididae) are among the main pests in lettuce and their control consists of frequent pesticide use. These aphids also attack species of several families of cultivated plants and weeds (Starý *et al.*, 2007) and are vectors of the lettuce-mosaic-virus (LMV), which results in leaf curl and affects plant growth and quality (Shawkat *et al.*, 1982).

The pest status of aphids depends, among others, on certain temperature conditions which result in fast development and strong population growth (Barbagallo *et al.*, 1998; Dixon, 1984). The developmental rate generally shows linearity between the lower and upper thermal limits (Campbell *et al.*, 1974) and is often used to estimate the lower temperature threshold and the degree-days needed for a species to complete its development (Lamb, 1961). Thus, studies about the influence of temperature on biological parameters of aphids help with understanding their development and population

growth, and are of great importance to the development and implementation of biological control programs (Ro *et al.*, 1998; Cividanes, 2003). The objective of this study was to determine the influence of temperature on the developmental time and survival of the aphids *A. solani*, *M. euphorbiae* and *U. ambrosiae*. With the results obtained during this study, their thermal requirements were estimated.

Materials and methods

Experiments were performed in the Laboratory of Biological Control of the Department of Entomology, Federal University of Lavras, Lavras, Minas Gerais, Brazil.

Collection and rearing of aphids

Lettuce plants, cultivar 'Veronica', containing colonies of *A. solani*, *M. euphorbiae* and *U. ambrosiae* were collected in a plastic tunnel with hydroponic lettuce production and transferred to the laboratory. After identification of the species, based on descriptions of Peña-Martines (1992), about 50 individuals of each aphid species were transferred with a paint brush to a rearing container. When mummies (i.e. parasitized aphids) were discovered, they were removed from the rearing container. Each container constituted a Petri dish (15 cm diameter) containing a lettuce leaf disc (14 cm diameter) on a layer of solidified 1% agar solution. The dishes were maintained in a climate chamber at 22 ± 1 °C, 70 ± 10% RH, and 12 h photophase. The leaf discs originated from pesticide-free plants that had been cultivated hydroponically. To avoid the presence of the contaminants, the leaf discs were cleaned by immersion in a solution of sodium hypochlorite 1% for about five

minutes, rinsed in water and then immersed in distilled water for about ten minutes (Diniz *et al.*, 2006). At the first signals of chlorosis or dehydration of the leaf disc, aphid colonies were transferred with a paint brush to another Petri dish containing a new lettuce leaf disc.

Development and thermal requirements

For each aphid species, 60 adult females obtained from the rearing procedure as described above were isolated individually in a Petri dish (10 cm diameter) containing a lettuce leaf disc (cultivar ‘Veronica’; 9 cm in diameter) and 1% agar layer, where they remained for a period of six hours. Then, the adult was removed and only one of the nymphs generated during this period was kept in each Petri dish. Tests were conducted in climate chambers at 16, 19, 22, 25, 28 and 31 ± 1 °C, 70 ± 10% RH and 12 h photophase. The aphids were moved to another Petri dish that contained a new lettuce leaf disc every four days. Development of the aphids was observed every 24 hours under a stereomicroscope and the developmental time and survivorship were recorded. The lower temperature threshold (T_b) and thermal constants (K) were determined with the hyperbole method (see section below).

Statistical analyses

The experiment was conducted in a completely randomized design with 60 replicates for each aphid species at each temperature (16, 19, 22, 25, 28 and 31 °C). Regression analyses (R Developmental Core Team, 2009) were applied to compare the development time and mortality of each aphid species at different temperatures. The averages and standard errors of nymphal developmental times were computed from the Kaplan-Meier estimator (Kaplan and Meijer, 1958). Pairwise tests of treatments at different temperatures were performed with the Log-rank test and an overall significance of 5% was maintained using a modified Bonferroni procedure (Bland and Altman, 2004). The relationships between total immature developmental time and temperature for the three aphid species were estimated by means of fitting with polynomial functions. To calculate the lower temperature threshold for devel-

opment and the thermal constant, we used the hyperbole method, which is based on the use of linear regression $Y = a + bX$, where Y is the reciprocal of the development time in days and X is the temperature in degrees Celsius (Silveira Neto, 1976; Campbell *et al.*, 1974, Haddad and Parra, 1984). To estimate the lower temperature threshold with this method, at least four temperatures are needed in the range of potential insect development (Campbell *et al.*, 1974).

Results and discussion

Developmental time of nymphal stages

The longest developmental times for first, second and fourth nymphal stages of *A. solani*, were found at 16 °C, and were 2.4, 2.5 and 3.4 days, respectively (figure 1). The longest developmental time for the third nymphal stage, which were found at 16 and 28 °C (2.4 and 2.7 days, respectively), did not differ significantly ($p < 0.05$) (figure 1). There was no difference in the duration of all nymphal stages of *A. solani* between 25 and 28 °C ($p > 0.05$) (figure 1). Even in the third nymphal stage, where the developmental time seems longer at 28 °C, there was no significant difference ($p > 0.05$) between the temperatures of 25 and 28 °C (1.6 and 2.7 days, respectively). At 31 °C little or no development took place, so the data are not presented.

The longest developmental time of the first nymphal stage of *M. euphorbiae* was observed at 16 °C (2.5 days). At 22, 25 and 28 °C the developmental times were 2.1, 1.7 and 1.6 days, respectively, and no significant differences in developmental time were found (figure 2). The developmental times of the second, third and fourth nymphal stages were similar at temperatures of 22 and 25 °C, and they were shorter than those found at 28 °C (figure 2). The fact that developmental times of the second, third and fourth nymphal stages were longer at 28 °C (2.3, 2.7 and 3.1 days, respectively) than at 22 °C (1.3, 1.4 and 2.5 days, respectively) and 25 °C (1.6, 1.4 and 2.3 days, respectively), and that no development took place at 31 °C, demonstrates that high temperatures have a negative effect on the development of *M. euphorbiae*.

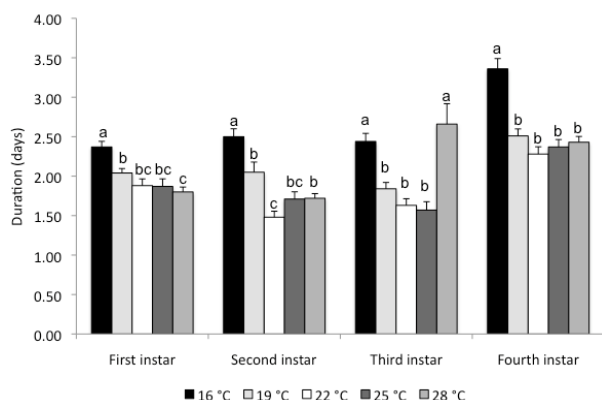


Figure 1. Developmental time of nymphal stages of *A. solani* at different temperatures. For each nymphal stage, bars with the same letters are not significantly different at $P < 0.05$ (log-rank test).

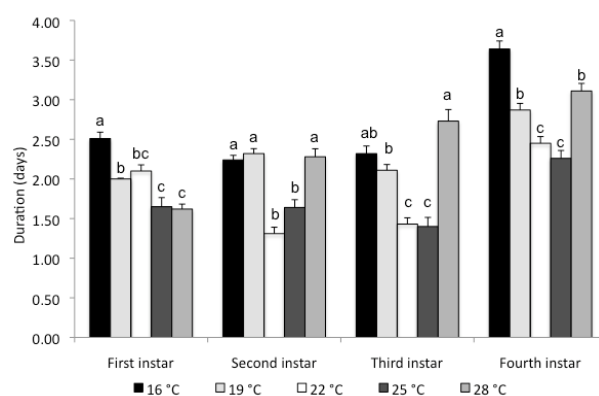


Figure 2. Developmental time of nymphal stages of *M. euphorbiae* at different temperatures. For each nymphal stage, bars with the same letters are not significantly different at $P < 0.05$ (log-rank test).

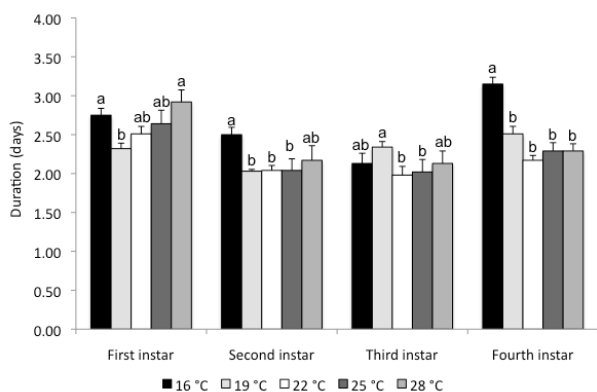


Figure 3. Developmental time of nymphal stages of *U. ambrosiae* at different temperatures. For each nymphal stage, bars with the same letters are not significantly different at $P < 0.05$ (log-rank test).

The shortest developmental time of the first nymphal stage of *U. ambrosiae* occurred at 19 °C (2.3 days), while at the other temperatures higher values were observed (figure 3). The developmental time of the second nymphal stage was longer at 16 °C (2.5 days) when compared to those at 19, 22 and 25 °C (figure 3). No significant differences were found for the developmental times observed at all temperatures for the third nymphal stage of *U. ambrosiae*. The fourth nymphal stage showed the longest developmental time at 16 °C (3.2 days); at the other temperatures (19, 22, 25 and 28 °C) no significant differences were found in developmental times of the fourth stage (figure 3). At 31 °C little or no development took place, so the data are not presented. The similarity in developmental times of nymphal stages for *U. ambrosiae* in the temperature range of 19-28 °C (figure 3) indicates that this species might be sensitive to higher temperatures than 19 °C as developmental times do no longer significantly decrease at temperatures above 19 °C.

Total immature developmental time

The relationship of total immature developmental time and temperature for *A. solani* and *U. ambrosiae* can be represented best by means of a polynomial function of the second order (figure 4). The estimated values obtained with the equations indicate a decreasing period of development between 16 and 25.7 °C for *A. solani* and 16 to 26.9 °C for *U. ambrosiae*. At higher temperatures than 25.7 °C for *A. solani* and 26.9 °C for *U. ambrosiae* an increasing developmental period was estimated (figure 4), although a small reduction in developmental times of the two species was observed in the experiments. The fact that developmental times do no longer decrease above a certain temperature is the first indication that these higher temperatures are non-optimal for the development of an insect (Gilbert and Raworth, 1996; Huey and Berrigan, 2001). When we apply this reasoning to the observed developmental times of *A. solani* and *U. ambrosiae*, we may conclude that temperatures above 22 °C - where developmental time no longer decreases - may have a negatively influence on the development of these two aphid species.

The estimated developmental time of *M. euphorbiae* followed a third order polynomial function: it decreased between 16 and 25.7 °C and clearly increased between 25.7 and 28 °C (figure 4). These results indicate adverse temperature effects on the development of this species above 25.7 °C.

Similar developmental times were reported for other common aphids in lettuce, like for *Myzus persicae* (Sulzer) (Chagas Filho *et al.*, 2005) and for *Nasonovia ribis-nigri* (Mosley) (Diaz and Fereres, 2005). Auad *et al.* (2002) observed that *U. ambrosiae* had developmental times of 16.0, 8.5 and 7.3 days at 15, 20 and 25 °C, respectively, and these are in the same range as our data.

Lower temperature threshold (T_b) and thermal constant (K)

The lower temperature thresholds or base temperatures determined for *A. solani* and *M. euphorbiae* were 1.09 and 1.05 °C, respectively, and the thermal constants (K) were 142.9 and 144.9 degree days (DD), respectively (figures 5 and 6). Due to the levelling of the rate of development in *U. ambrosiae* it was not possible to estimate the lower temperature threshold for this species with the hyperbole method that we used (Silveira Neto, 1976; Campbell *et al.*, 1974; Haddad and Parra, 1984) (figure 7).

It is known that the thermal threshold and thermal constant are closely related to the adaptation of populations to the local climate (Blackman, 1987; Ikemoto, 2003), so populations of different species which occur under the same climate conditions often have similar thermal requirements, like we found for *A. solani* and *M. euphorbiae*. The lower temperature threshold can be used as a good indicator of the adaption of populations to certain climate conditions. Also Turak *et al.* (1998) found very similar values of T_b and K for two populations of *Sitobion miscanthi* (Takahashi) and *Sitobion near fragariae*, species belonging to the tribe Macrosiphini, like *M. euphorbiae*. The opposite may happen

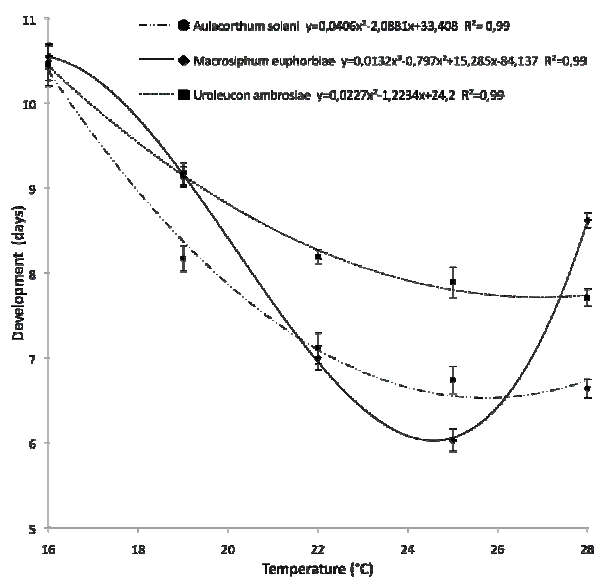


Figure 4. Developmental time of nymphal stages of *A. solani*, *M. euphorbiae* and *U. ambrosiae* as a function of temperature.

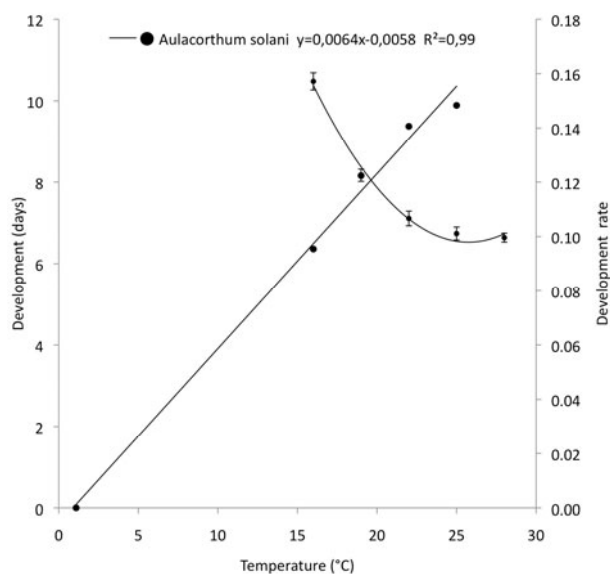


Figure 5. Immature developmental rate of *A. solani* as a function of temperature.

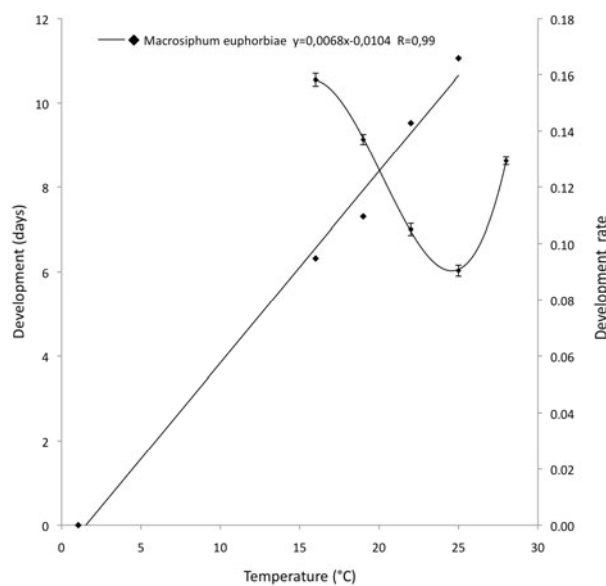


Figure 6. Immature developmental rate of *M. euphorbiae* as a function of temperature.

when populations of the same species are exposed to different climate conditions, resulting in different values for the lower temperature threshold and thermal constant (Royer *et al.*, 2001; Sampaio *et al.*, 2003)

The low T_b values of about 1 °C estimated for *A. solani* and *M. euphorbiae*, are indicating adaptation of these species to low temperatures (figures 5 and 6). In the field, those low T_b values make it possible for these species to start population growth when it is still relatively cold (Auaud *et al.*, 2002; Stary *et al.*, 2007).

Immature survival

Survival rates of immatures at 16, 19, 22, 25 and 28 °C of *A. solani* were 90, 92.5, 90, 61.7 and 55%, respectively (figure 8). For *M. euphorbiae*, survival rates were 83.3, 83.3, 85.0, 71.7, and 62.0%, respectively. For *U. ambrosiae* survival rates were 86.7, 95.0, 78.3, 65.0 and 36.7%, respectively. Immature survival of *A. solani*, *M. euphorbiae* and *U. ambrosiae* was strongly influenced by temperature, and was particularly low at 25 and 28 °C, while no survival at all was observed at 31 °C (figure 8).

The survival data were fit to a polynomial function of the second order (figure 8) and the fitted curves initially show a slight increase in survivorship followed by a decrease. According to the fitted curves estimated survival was highest at 17.2, 18.7 and 18.1 °C for *A. solani*, *M. euphorbiae* and *U. ambrosiae*, respectively.

The developmental times (figure 4) as well as the survival rates (figure 8) of *A. solani*, *M. euphorbiae* and *U. ambrosiae* indicate negative effects of high temperatures for these species. At temperatures around 22 °C the highest survival rates and shortest developmental times were observed for *A. solani* and *M. euphorbiae*. As for *U. ambrosiae* the optimal combination of survival rate and developmental time was observed at 19 °C. Studies by Chagas *et al.* (2005) and Diaz and Fereres (2005) showed that survival rates were highest at 15 and 20 °C for the aphid *M. persicae*, and between 16 and

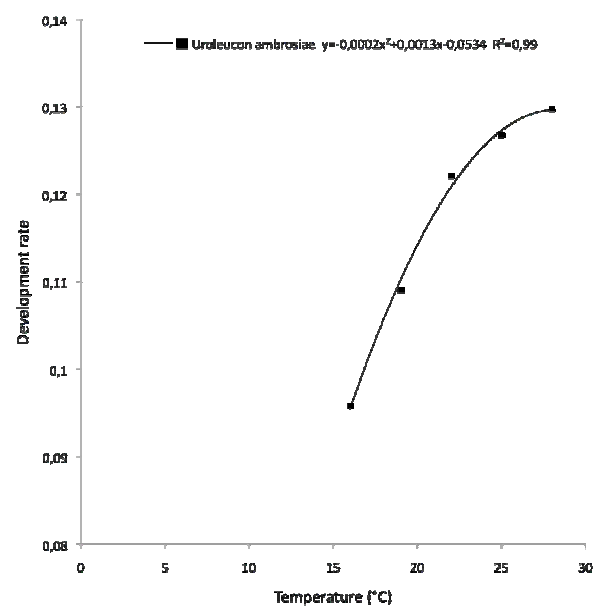


Figure 7. Immature developmental rate of *U. ambrosiae* as a function of temperature.

24 °C for *N. ribisnigri*, while the highest mortality for both species occurred at temperatures around 30 °C.

In this study, the earlier described negative effects observed at 28 °C and 31 °C, indicate that these species of aphids are not adapted to those high temperatures. In our study location (Lavras, Minas Gerais, Brazil, 21°14'43"S, 44°59'59"W), the occurrence of *A. solani*, *M. euphorbiae*, *U. ambrosiae* was recorded only between April and October (Stary *et al.*, 2007), during which the average maximum temperature usually does not exceed 26.9 °C (Brasil, 1992). Thus, the insects used in this study will only temporarily experience temperatures exceeding 26.9 °C during the period of April - October in their natural environment at our location.

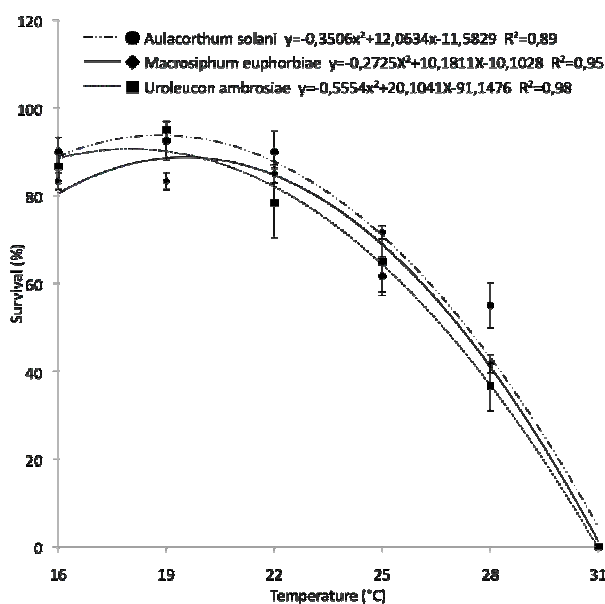


Figure 8. Immature survival of *A. solani*, *M. euphorbiae* and *U. ambrosiae* as a function of temperature

Conclusions

The results of this study indicate that *A. solani*, *M. euphorbiae* and *U. ambrosiae* are well adapted to moderately high temperatures in the range of 16-25 °C. The optimal temperatures for development and survival are 19 °C for *U. ambrosiae*, and 22 °C for *A. solani* and *M. euphorbiae*. Our results can be used in the selection of natural enemies for biological control of these species (De Conti *et al.*, 2011).

Acknowledgements

The authors wish to thank the Coordenação de Aperfeiçoamento do Ensino Superior (CAPES) and the Fundação de Amparo a Pesquisa do Estado de Minas Gerais (FAPEMIG) for financial support. Comments of the three reviewers were highly appreciated and helped us to improve the paper.

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Received June 16, 2010. Accepted March 1, 2011.