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# Performance of immatures of three Neotropical Miridae at five different temperatures, reared on *Ephestia kuehniella* eggs on tobacco plants

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## Abstract

Effects of temperature (16, 20, 24, 28 and 32 ± 1 °C), host plant (*Nicotiana tabacum* L.) and factitious prey (eggs of *Ephestia kuehniella* Zeller) on immature development of three recently found Neotropical mirids, *Campyloneuropsis infumatus* (Carvalho), *Engytatus varians* (Distant) and *Macrolophus basicornis* (Stal) were studied at RH 70 ± 10% and 12h photophase in climate cabinets. These mirids are being evaluated for biological control of the South American tomato borer *Tuta absoluta* (Meyrick) and other pests on tomato. Survival of eggs of the three mirid species on tobacco was high (> 80%) at 16-28 °C, but lower (< 80%) at 32 °C. Development times decreased with increasing temperature from 16-28 °C. Nymphal survival was higher (84-96%) at 20, 24 and 28 °C than at 16 and 32 °C (46-83%). The sex ratio of *C. infumatus* was strongly female biased at all temperatures, whereas it was 1:1 for the other two species. The lower temperature thresholds for egg-adult development of *C. infumatus*, *E. varians* and *M. basicornis* were 9.4, 9.4 and 7.9 °C, and their thermal constants were 384.6, 384.6 and 476.2 DD, respectively. Temperatures between 24 to 28 °C are best for immature performance and for rearing of these mirids species. Eggs of the factitious host *E. kuehniella* provide adequate food for their mass production. Optimal temperatures for best mirid predator performance are similar to those for the pest *T. absoluta*, indicating good climate matching.

**Key words:** *Campyloneuropsis infumatus*, *Engytatus varians*, *Macrolophus basicornis*, *Tuta absoluta*, biological control, mass production, thermal constants.

## Introduction

Tomato *Solanum lycopersicum* L. (Solanaceae) is the most widely produced fruit vegetable in the world and also plays an important role in the agricultural economy of Brazil, one of the main tomato producers worldwide (Agrianual, 2016). Tomato can be attacked by various insect pests, including the South American tomato borer *Tuta absoluta* (Meyrick) (Lepidoptera Gelechiidae), which may cause complete crop loss without pest control (Guedes and Picanço, 2012). In Brazil, *T. absoluta* is controlled by frequent pesticide sprays (Thomazini *et al.*, 2001) and as a consequence, Brazilian tomato borer populations have acquired resistance to several active ingredients, resulting in a so-named pesticide treadmill, decimation of natural enemies and high residue levels on the tomato fruit (van den Bosch, 1978; Siqueira *et al.*, 2000; 2001; Silva *et al.*, 2011; Guedes and Picanço, 2012). The rapid development of resistance to frequently applied pesticides necessitates a search for alternative control methods, such as biological control.

Heteropteran predators, particularly those belonging to the family Miridae (Calvo *et al.*, 2012; van Lenteren, 2012; Jaworski *et al.*, 2013; Pazyuk *et al.*, 2014; van Lenteren *et al.*, 2018a) might provide control of *T. absoluta*, while these polyphagous predators may also be used for control of other key pests of tomato (Calvo *et al.*, 2009; Martínez *et al.*, 2015; Pérez-Hedo *et al*, 2015). In Spain, for example, the Palearctic mirid *Macrolophus pygmaeus* (Rambur) and Paleaotropic *Nesidiocoris tenuis* (Reuter) (Hemiptera Miridae) are effectively controlling *T. absoluta* and *Bemisia tabaci* (Gennadius) (Calvo *et al.*, 2012). In Brazil, the species *Campyloneuropsis infumatus* (Carvalho), *Engytatus varians* (Distant) and *Macrolophus basicornis* (Stal) (Hemiptera Miridae) were recently found in the laboratory and field to prey on eggs and larvae of *T. absoluta* (Bueno *et al.*, 2013a; 2013b; van Lenteren *et al.*, 2017), nymphs of *B. tabaci* and eggs and larvae of various other lepidopteran pests of tomato (Bueno *et al.* 2013a). Bueno *et al.* (2012; 2013a; 2013b) and Silva *et al.* (2016) reported that these Neotropical mirids can use tomato plants as an oviposition substrate, can complete their development on it, and, contrary to other predators, like *Geocoris punctipes* (Say) (Hemiptera Geocoridae) and *Orius insidiosus* (Say) (Hemiptera Anthocoridae), can easily walk on the stems of tomato plants despite the presence of sticky and poisonous trichomes (Bueno *et al.*, 2013a). Predatory Miridae show the characteristic of zoophytophagy: in addition to eat animal prey, they also feed on plant tissues to complement or supplement nutritional needs or as a source of water (Wheeler, 2001; Albajes and Alomar, 2008; Bueno and van Lenteren, 2012). Phytophagy can result in both beneficial and detrimental effects. Beneficial because plant feeding allows them to survive periods of low pest abundance, detrimental because it may result in plant injury and yield loss. The two mirids commercially used for biological control of important pests in greenhouse tomatoes, *M. pygmaeus* and *N. tenuis*, may cause serious plant injury under specific conditions when prey availability is low and predator density is high. Particularly *N. tenuis* needs special attention with regard to population management at high densities in combination with low pest populations, because extensive plant feeding by this predator results in necrotic rings on the stems, shoots, leaf petioles and flower stalks, causing abortion of flowers and young fruits, reduced growth of tomato plants and yield loss (Calvo *et al*., 2009). However, when properly managed, the European mirids are considered important biocontrol agents in tomato IPM and are used on a large scale in the Meditarranean region (Pérez-Hedo and Urbaneja, 2016). Remarkably, nymphs and adults of the three zoophytophagous Neotropical mirids appeared to cause little injury to tomato seedlings and fruit, even when present in high densities and in the absence of prey in the laboratory (Silva *et al*., 2017a) and in presence of prey in the greenhouse (van Lenteren *et al*., 2018b).

However, the potential of these Neotropical mirids to control pest populations of *T. absoluta* in the field still remains to be demonstrated. Knowledge about the predator's activity at a range of temperatures occurring in the field and in greenhouses in tomato production areas of Brazil is important to be able to determine their potential as natural enemies for control of *T. absoluta* and other pests on tomato. Also, information about their lower temperature threshold and thermal constant assists in determining their periods of activity in the field. Hughes *et al*. (2010) reported that a predator would have a selective advantage over its prey if it has a lower temperature threshold and a smaller thermal constant than its prey, because it would have a greater number of generations per year and would be active over a wider range of temperatures than the prey. On the other hand, synchronization of the life history of the predator to that of its prey is important at a range of local tomato production climate conditions (van Lenteren, 2010; Clarke, 2017). Interestingly, Horn (1998) found that biological control agents often have optimal development temperatures that are different from their prey.

Mass rearing of natural enemies is a critical step in a biological control programme. Thus, information about responses to different temperatures and thermal requirements of these Brazilian mirid predators can also assist in designing an efficient mass rearing system and in estimating the success of establishment after release in tomato crops. Further, we were interested in the quality of eggs of *Ephestia kuehniella* Zeller (Lepidoptera Pyralidae) as a factitious prey for these predators, because mass rearing of these predators is much easier and cheaper on this factitious prey than, for example, on the pest *T. absoluta* (Mollá *et al*., 2014). We are aware that several other factitious prey species, either alone or in combination with artificial media, have been tested to rear other mirid species (e.g. Vandekerkhove *et al*., 2011; Aubrey *at al*., 2015), but we have limited our first experiments to the easily available, often used and well known flour moth *E. kuehniella*.

In this paper, data are provided for the development time and survival of eggs and nymphs, weight of the 4th and 5th nymphal instars, egg-adult development time, lower developmental thresholds and thermal constants for egg-adult development, and sex ratio of emerged adults of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures (16, 20, 24, 28 and 32 ± 1 °C) with *E. kuehniella* eggs as food source and with tobacco as host plant. This temperature range was selected based on the optimum temperatures for tomato production in Brazil (Naika *et al*., 2006), though lower and higher temperatures may occasionally occur. Both, growers producing tomatoes in the field and in greenhouses try to adhere at these optimal temperature schedules.

## Materials and methods

### Rearing of the mirid predators

Adult mirids (*C. infumatus*, *E. varians* and *M. basicornis*) were collected in areas cultivated with tomato (*S. lycopersicum*) and tobacco *Nicotiana tabacum* L. (Solanaceae) (Bueno *et al*., 2012). Subsequently, a rearing colony was set up in the laboratory as described in Bueno *et al*. (2013a). Tobacco plants, *N. tabacum* cv TNN were grown in a greenhouse on organic substrate (75% *Pinus* rusk and 25% vermiculite). Seedlings with two pairs of leaves were transplanted to 2 L plastic pots and maintained until attaining a height of 25 cm. Field collected adults of each predator species were individually released in acrylic cages (60 × 30 × 30 cm) containing a tobacco plant as oviposition substrate and a water source. Tobacco plants and the adult predators remained in the cages for seven days. Then, the plants containing mirid eggs were transferred to new cages. Eggs of *E. kuehniella* were offered *ad libitum* as food to the mirid nymphs and adults twice weekly. The stock rearings were kept in a climate room at 25 ± 1 °C, RH 70 ± 10% and 12 h photophase.

### Development, survival and morphological aspects of eggs

Ten pairs of each predator species were released in glass pots (1.7 L) containing a plastic cup (200 mL) (8.5 cm diameter × 5.5 cm height) with a tobacco seedling with two pairs of leaves as oviposition substrate and *E. kuehniella* eggs as food *ad libitum* for a 24h period. The glass pots were sealed with voile fabric and kept in a climate room at 25 ± 1 °C, RH 70 ± 10% and 12 h photophase. After 24 h, the number of eggs was counted using a stereomicroscope (40×). Eggs on tobacco leaves, petioles and stems could be seen by putting the seedling on a light source, which renders the eggs darker than the plant tissue, and by looking for opercula which protrude from the plant tissue (Bueno *et al*., 2013a). Subsequently, the roots of the seedlings were wrapped in moistened cotton in order to avoid wilting and transferred to Petri dishes (15 cm) sealed with PVC film. For each predator species and each temperature (16, 20, 24, 28 and 32 ± 1 °C), 10 Petri dishes with a seedling containing eggs were put in climate chambers with a RH of 70 ± 10% and a 12 h photophase. Egg development time and egg survival, expressed by the percentage of 1st instar nymphs hatching, was determined for each predator species. To be able to study morphological characteristics, eggs of *C. infu-*

*matus*, *E. varians* and *M. basicornis* were removed from the longitudinal veins of the leaves of tobacco seedlings with the aid of stylus and observed using a stereomicroscope (30×) (Wheeler, 2001).

### Development, survival, size of nymphs, sex ratio at the adult emergence, and weight of 4th and 5th instar nymphs

To obtain newly-hatched nymphs, potted *N. tabacum* plants 25 cm in height were kept in acrylic cages (60 × 30 × 30 cm) along with 100 females and 100 males of each adult predator species for a period of 24 h in climate chambers at 16, 20, 24, 28 and 32 ± 1 °C, RH 70 ± 10% and 12 h photophase. Next, plants containing eggs were transferred to new acrylic cages and were kept at the same conditions. These plants were observed daily at the same time for hatched nymphs. One hundred newly-hatched nymphs of each predator species were individually placed in Petri dishes (5 cm diameter) containing *N. tabacum* leaf discs (4.5 cm diameter) on 1% agar-water layer and *E. kuehniella* eggs *ad libitum*. Each Petri dish was sealed with a paper towel secured by a rubber band to prevent escape of the nymphs. The leaf discs were changed twice a week, and at the same time eggs of *E. kuehniella* were supplied *ad libitum*. Each 10 nymphs were considered a replicate in a total of 10 replicates for data analysis. Instar changes and nymphal mortality were observed daily at the same time using a stereomicroscope (30×). For each temperature, the development time of each instar, total nymphal development, egg-adult development, nymphal survival and sex ratio of the adults at emergence were determined. The weight of 20 individuals of the 4th and 5th nymphal instars was determined by individually placing nymphs in plastic tubes on an analytical precision scale (d-0.0001g) (Shimadzu, AW 220). Nymphal instar characteristics as size (measured by the largest width of the cephalic capsule between the outer margins of the compound eyes using a micrometer reticule coupled in stereomicroscope, 40×) and body colour (observed by stereomicroscope at 30×) were recorded.

### Data analysis

Data for cohorts of 100 nymphs (10 nymphs per replicate) were used to determine the development time of each instar, total nymphal development time, nymphal survival and sex ratio of the adults at emergence for each predator species at each temperature. All statistical procedures were performed using the software R 3.2.0 (R Development Core Team, 2015). First, the data were subjected to an exploratory analysis to verify the residual normality with the Shapiro-Wilk test, and then the data were tested for residual homogeneity with the Levene test (Gujarati, 2004); both tests were performed at P = 0.01 and P = 0.05. Variables with residuals not normally distributed and/or heteroscedastics (development of eggs, total development of nymphs and egg to adult development, survival of nymphs and weight of the 4th and 5th instars of *E. varians* and the 4th instar of *M. basicornis*) were analyzed with Generalized Linear Models (Nelder and Wedderburn, 1972; McCullagh and Nelder, 1989; Lee *et al*., 2006).

The data for egg and nymphal development and egg to adult development were adjusted to Poisson distributions with a logarithmic link function. The other data (survival of nymphs and weight of the 4th and 5th instars of *E. varians* and the 4th instar of *M. basicornis*) were adjusted to Quasi-Poisson distributions with a logarithmic link function. The Wald $\chi^2$ test was used to test for significance of the effects of the models and the parameters of the regression analyses.

We analyzed only the effect of temperature on weight of the 4th and 5th instars of *E. varians* and the 4th instar of *M. basicornis*, but we considered temperature effect, species of mirid predator and their interaction as fixed factors for the other analyzed data (egg, nymphal and egg to adult development and survival of nymphs). Regression analysis was used for evaluation of the temperature effect on egg, nymphal and egg to adult development and survival of nymphs. The Dunn test was used to compare means of egg to adult development time and survival of nymphs among the three predator species for each temperature, and to compare weight of the 4th and 5th instars of *E. varians* and the 4th instar of *M. basicornis* among temperatures.

Variables that met the assumptions of normality and homogeneity were analyzed by ANOVA (survival of eggs, weight of 4th and 5th instars of *C. infumatus* and weight of 5th instar of *M. basicornis*). Temperature, species of mirid predator and their interaction were included as independent variables and analyzed by a two-way ANOVA for egg survival. Data were transformed according to $y = \arcsin \sqrt{(x / 100)}$ and regression analysis was used to analyze the effect of temperature on egg survival. A one-way ANOVA and a Tukey test were applied to analyze the effect of temperature on the weight of 4th and 5th instars of *C. infumatus* and weight of 5th instar of *M. basicornis*. Data of the 4th instar of *C. infumatus* and the 5th instar of *M. basicornis* were transformed according to $y=\sqrt{x}$. To determine deviations from a 1:1 male/female sex ratio, the data were analyzed by a homogeneity $\chi^2$ test. For all analyses, a P = 0.05 was used as significance level.

The lower temperature thresholds for development (LDT) and the thermal constants (K) of the three mirid species were calculated with the hyperbole method. This method makes use of a linear regression $y$=a+b$x$, where $y$ is the reciprocal of the development time in days and $x$ is the temperature in degrees Celsius (Campbell *et al*., 1974; Bergant and Trdan, 2006). To estimate the lower temperature threshold with this method, measurements are needed for at least four different temperatures in the range of insect development (Campbell *et al*., 1974), and as we have data for five temperatures, we have met this prerequisite.

## Results

### Morphology, development time and survival of eggs

Females of *C. infumatus*, *E. varians* and *M. basicornis* oviposit in the main midrib of the leaves, and rarely in the petioles and stems of tobacco plants. The eggs are inserted into the plant tissue with only their opercula visible. Eggs of *C. infumatus* are elongated and slightly curved, 0.762 mm long and 0.254 mm wide, milky-white to translucent with a smooth outer egg surface and two short respiratory extensions (spiracles), one on the concave region and another on the convex region near the operculum. Eggs of *E. varians* are 0.915 mm long and 0.254 wide, are milky-white to pale yellow and have only one spiracle in the concave region near the operculum, which is longer than those of *C. infumatus* eggs. Eggs of *M. basicornis* are similar to those of *E. varians*, 0.813 mm long and 0.254 mm wide, and milky-white to translucent in colour.

No significant interaction was found between temperature effect and species of mirid for the development time of eggs (P = 0.5457, df = 8; 135). However, temperature significantly affected the development time of eggs (P < 0.0001, df = 4; 135) of the three mirids: development time decreases with increasing temperature (figure 1). Development times of the eggs of the three mirids did not significantly differ at each test temperature (P = 0.0549, df = 2; 135; table 1). Also, no significant interaction was found between temperature effect and species of mirid for egg survival ($F_{8;135}$ = 0.0840, P = 0.9438). Further, no significant differences were found for egg survival ($F_{2;135}$ = 0.351, P = 0.9185) of the three mirid species at each temperature. But temperature significantly affected egg survival ($F_{4;135}$ = 15.763, P < 0.0001) of the mirids, and egg survival was highest at the intermediate temperatures (figure 2).

Because no statistical differences existed between species at a certain temperature in egg development time, egg survival and development time of nymphs (table 1), we have combined the data of the three species to calculate the relationships represented in figures 1, 2 and 3.

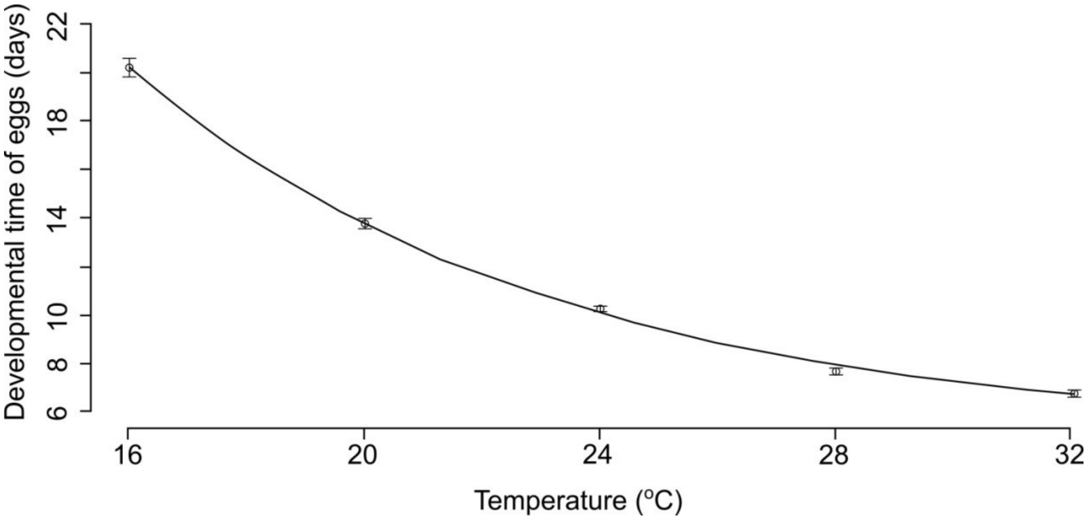

**Figure 1.** Developmental time of eggs in days (means ± SE) of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase. $f(x)$=exp(5.2862259-0.1792171x+0.0022983x$^2$).

**Table 1.** Developmental time in days (means ± SE) of eggs, nymphal instars, total nymphal development (TND) and egg-adult development of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase; n = number of individuals.

| T °C | Species | Eggs* | n | Nymphal instars* | | | | | | | | | | TND* | Egg-adult** | n |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | 1st | n | 2nd | n | 3rd | n | 4th | n | 5th | n | | | |
| 16 | *C. infumatus* | 18.5±1.37 | 400 | 6.3±0.13 | 95 | 7.2±0.17 | 89 | 7.6±0.21 | 81 | 8.3±0.10 | 78 | 12.8±0.19 | 64 | 41.7±0.92 | 60.2±0.77A | 64 |
| | *E. varians* | 19.3±0.91 | 498 | 6.5±0.13 | 95 | 6.4±0.15 | 81 | 7.4±0.19 | 73 | 9.1±0.18 | 68 | 13.3±0.31 | 51 | 42.0±1.15 | 61.3±1.21A | 51 |
| | *M. basicornis* | 22.6±1.10 | 258 | 7.3±0.11 | 97 | 6.5±0.14 | 92 | 7.1±0.16 | 87 | 8.0±0.16 | 74 | 12.2±0.24 | 71 | 39.0±2.04 | 61.6±2.22A | 71 |
| 20 | *C. infumatus* | 13.0±1.16 | 190 | 4.0±0.72 | 100 | 3.3±0.05 | 99 | 3.4±0.06 | 98 | 3.7±0.08 | 97 | 6.3±0.05 | 93 | 20.3±0.15 | 33,3±0.31B | 93 |
| | *E. varians* | 13.6±1.01 | 218 | 4.4±0.56 | 98 | 3.3±0.05 | 97 | 3.4±0.06 | 91 | 3.8±0.04 | 91 | 6.0±0.03 | 89 | 20.9±0.18 | 34.5±0.40B | 89 |
| | *M. basicornis* | 14.6±0.91 | 213 | 5.5±0.67 | 100 | 3.7±0.05 | 100 | 3.8±0.04 | 100 | 4.3±0.05 | 97 | 7.2±0.05 | 92 | 24.7±0.33 | 39.3±0.42A | 92 |
| 24 | *C. infumatus* | 9.9±0.42 | 250 | 3.2±0.04 | 100 | 2.3±0.05 | 100 | 2.5±0.05 | 97 | 3.0±0.07 | 97 | 4.4±0.06 | 96 | 16.1±0.23 | 26.0±0.29B | 96 |
| | *E. varians* | 10.0±0.40 | 132 | 3.6±0.05 | 99 | 2.5±0.07 | 96 | 2.5±0.05 | 93 | 3.2±0.06 | 90 | 4.7±0.05 | 84 | 17.2±0.59 | 27.2±0.63B | 84 |
| | *M. basicornis* | 10.7±0.21 | 322 | 3.2±0.04 | 100 | 3.3±0.05 | 97 | 3.3±0.06 | 96 | 3.5±0.07 | 94 | 5.2±0.04 | 93 | 18.2±0.20 | 28.9±0.20A | 93 |
| 28 | *C. infumatus* | 7.5±0.36 | 237 | 3.1±0.05 | 100 | 2.2±0.06 | 100 | 1.9±0.05 | 100 | 2.2±0.05 | 96 | 3.7±0.07 | 92 | 12.9±0.23 | 20.4±0.26B | 92 |
| | *E. varians* | 7.1±0.35 | 216 | 2.9±0.03 | 100 | 2.0±0.03 | 100 | 1.5±0.05 | 98 | 2.4±0.05 | 97 | 3.2±0.05 | 94 | 12.6±0.27 | 19.7±0.23B | 94 |
| | *M. basicornis* | 8.2±0.71 | 221 | 3.1±0.03 | 99 | 2.2±0.05 | 99 | 2.3±0.05 | 98 | 2.5±0.05 | 96 | 4.1±0.05 | 94 | 15.0±0.21 | 23.2±0.24A | 94 |
| 32 | *C. infumatus* | 7.0±0.51 | 185 | 3.3±0.06 | 94 | 1.9±0.05 | 89 | 1.9±0.04 | 83 | 2.6±0.07 | 74 | 3.7±0.09 | 45 | 13.3±0.37 | 20.4±0.36A | 45 |
| | *E. varians* | 6.1±0.14 | 352 | 2.1±0.03 | 100 | 1.8±0.03 | 99 | 1.9±0.02 | 92 | 2.0±0.04 | 87 | 3.2±0.07 | 51 | 11.2±0.20 | 17.3±0.22B | 51 |
| | *M. basicornis* | 7.2±0.21 | 146 | 2.8±0.03 | 98 | 1.9±0.02 | 96 | 2.0±0.02 | 92 | 2.3±0.05 | 91 | 3.8±0.05 | 83 | 13.1±0.18 | 20.3±0.20A | 83 |

*Means were not significantly different when comparing the species at the same temperature (Wald $\chi^2$ test; p > 0.05).

**Means followed by the same capital letters in columns are not significantly different when comparing the species at the same temperature (Dunn test; p > 0.05).

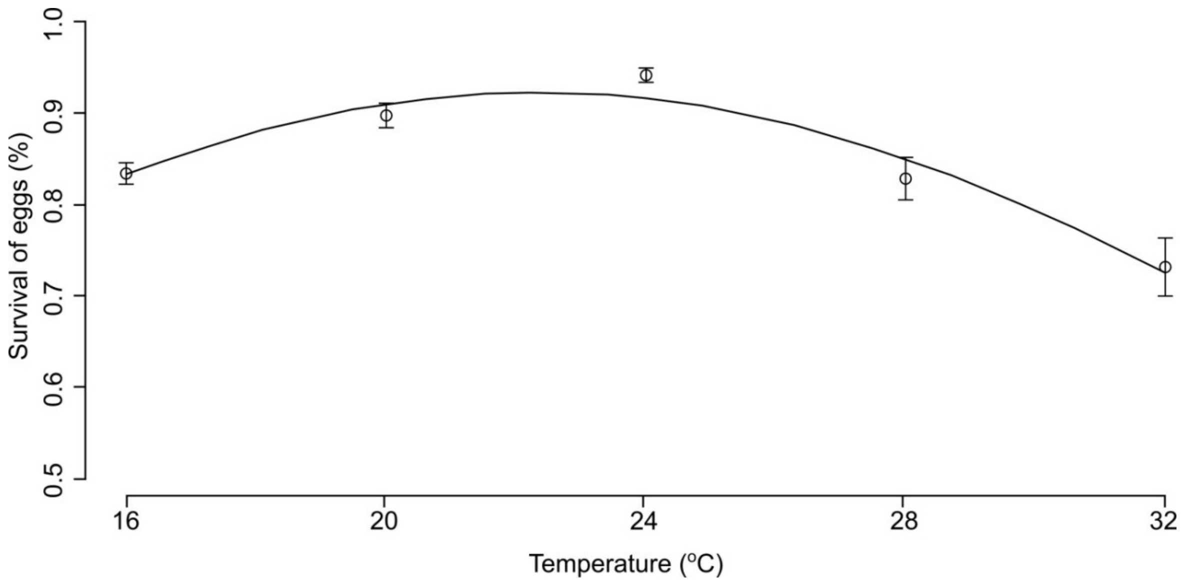

**Figure 2.** Fraction survival of eggs (means ± SE) of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase. *y*= $-0.2134x^2+9.5603x-15.045$, $R^2 = 0.9403$.

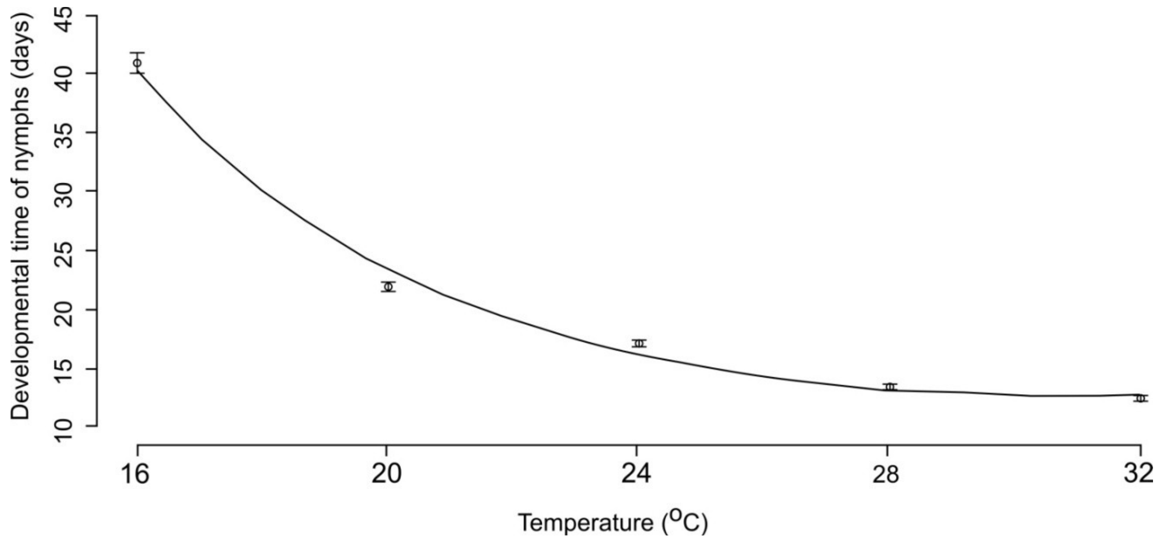

**Figure 3.** Developmental time of nymphs in days (means ± SE) of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase. *f(x)*=$\exp(7.5439642-0.3249259x+0.0052779x^2)$.

### Development time of instars, total nymphal development time, egg-adult development time, nymphal survival and thermal requirements

No significant interaction was found between temperature effect and species of mirid for total nymphal development time (P = 0.1733, df = 8; 135). Also, no significant differences were found for the development time of instars (P = 0.3358, df = 2; 135) of the three mirid species at each temperature. Temperature affected total nymphal development time (P < 0.0001, df = 4; 135), which decreased with increasing temperature from 16 to 28 °C and stabilized between 28 and 32 °C (figure 3). In addition, no significant interaction was found between temperature effect and species of mirid for egg-adult development (P = 0.2785, df = 8; 135). Egg-adult development was significantly different among the three mirid species at some temperatures (P = 0.0321, df = 2; 135). Egg-adult development of the three mirid species was not different at 16 °C (Dunn = 1.0245, df = 2, P = 0.6000), but development of *M. basicornis* was longer than that of *E. varians* at the other four temperatures, and longer than that of *C. infumatus* at 20 °C (Dunn = 21.1718, df = 2, P < 0.0001), 24 °C (Dunn = 14.8803, df = 2, P < 0.0001) and 28 °C (Dunn = 20.5157, df = 2, P < 0.0001) (table 1). The egg-adult development of *C. infumatus* was significantly longer than that of *E. varians* only at 32 °C (Dunn = 18.9966, df = 2, P < 0.0001) (table 1). As expected, temperature affected the egg-adult development (P < 0.0001, df = 4; 135) of the three mirids, showing shorter development with increasing temperature (figure 4).

A significant interaction was found between temperature effect and species of mirid for survival of the nymphs (P = 0.0049, df = 8; 135). The percentage nym-

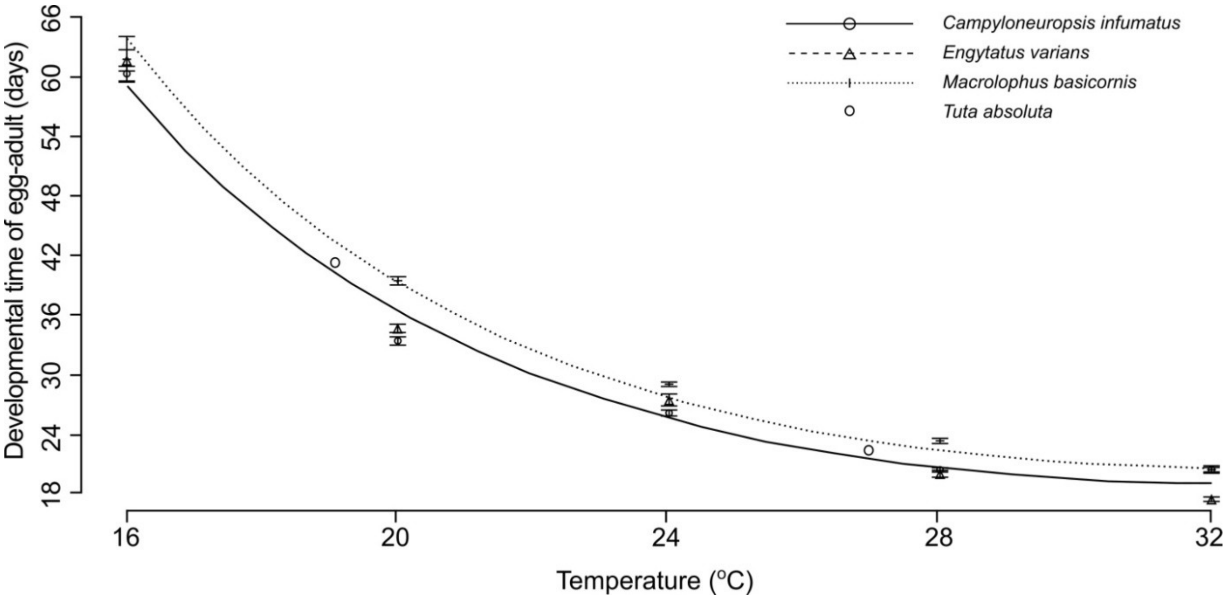

**Figure 4.** Developmental time egg-adult in days (means ± SE) of three species of Miridae at five temperatures, RH 70 ± 10% and 12h photophase. *C. infumatus* $f(x)=\exp(7.3645425-0.2729148x+0.0042136x^2)$, *E. varians* $f(x)=\exp(7.3645425-0.0010038-0.2729148x+0.0042136x^2)$; *M. basicornis* $f(x)=\exp(7.3645425+0.0779611-0.2729148x+0.0042136x^2)$. The dots represent egg-adult development time of the pest *T. absoluta* (data from Barrientos *et al.*,1998).

**Table 2.** Percentage survival of eggs and nymphs (means ± SE) and estimated total survival (%) of immatures of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase; n = number of individuals.

| T °C | Species | Survival (%) | | | | Estimated total survival of immatures*** |
|---|---|---|---|---|---|---|
| | | Eggs* | n | Nymphs** | n | |
| 16 | *C. infumatus* | 84.2 ± 1.81 | 400 | 64.0 ± 3.71AB | 64 | 53.8 |
| | *E. varians* | 83.5 ± 1.97 | 498 | 51.0 ± 4.06B | 51 | 42.5 |
| | *M. basicornis* | 82.3 ± 2.58 | 258 | 71.0 ± 4.33A | 71 | 58.4 |
| 20 | *C. infumatus* | 87.3 ± 2.53 | 190 | 93.0 ± 2.60A | 93 | 81.1 |
| | *E. varians* | 90.2 ± 2.34 | 218 | 89.0 ± 1.79A | 89 | 80.2 |
| | *M. basicornis* | 91.7 ± 2.05 | 213 | 92.0 ± 2.90A | 92 | 84.3 |
| 24 | *C. infumatus* | 95.7 ± 1.07 | 250 | 96.0 ± 2.21A | 96 | 91.8 |
| | *E. varians* | 92.3 ± 1.73 | 132 | 84.0 ± 4.26B | 84 | 77.5 |
| | *M. basicornis* | 94.3 ± 1.02 | 322 | 93.0 ± 2.13AB | 93 | 87.4 |
| 28 | *C. infumatus* | 84.2 ± 4.54 | 237 | 92.0 ± 2.49A | 92 | 77.4 |
| | *E. varians* | 82.5 ± 4.66 | 216 | 94.0 ± 2.21A | 94 | 77.5 |
| | *M. basicornis* | 81.8 ± 3.38 | 221 | 94.0 ± 2.21A | 94 | 76.8 |
| 32 | *C. infumatus* | 71.8 ± 6.62 | 185 | 46.0 ± 6.00B | 46 | 33.0 |
| | *E. varians* | 76.4 ± 4.92 | 352 | 51.0 ± 4.33B | 51 | 38.9 |
| | *M. basicornis* | 71.2 ± 5.10 | 146 | 83.0 ± 3.35A | 83 | 59.0 |

*Means were not significantly different when comparing the three species at the same temperature (F test; p > 0.05).

**Means followed by the same capital letters in columns are not significantly different when comparing species at the same temperature (Dunn test; p > 0.05).

***% estimated total immature survival (% eggs × % nymphs/100).

phal survival of *C. infumatus*, *E. varians* and *M. basicornis* (P < 0.001, df = 2; 135) was high (84-96%) and similar at 20 °C (Dunn = 2.3192, df = 2, p = 0.311), 24 °C (Dunn = 5.7068, df = 2, p = 0.060) and 28 °C (Dunn = 0.4554, df = 2, p = 0.800) (table 2). *C. infumatus* and *E. varians* showed lower survival than *M. basicornis* at 32 °C (Dunn = 17.6813, df = 2, p < 0.001), and *E. varians* showed lower survival (Dunn = 8.2576, df = 2, p = 0,020) than *C. infumatus* and *M. basicornis* at 16 °C (table 2, figure 5). The estimated total percentage survival of immatures of the three mirids was lowest at 32 °C (33-60%) and 16 °C (42-58%) and highest at 20, 24 and 28 °C (77-87%) (table 2).

The development rate of three mirid species as a function of temperature is presented in table 3. The

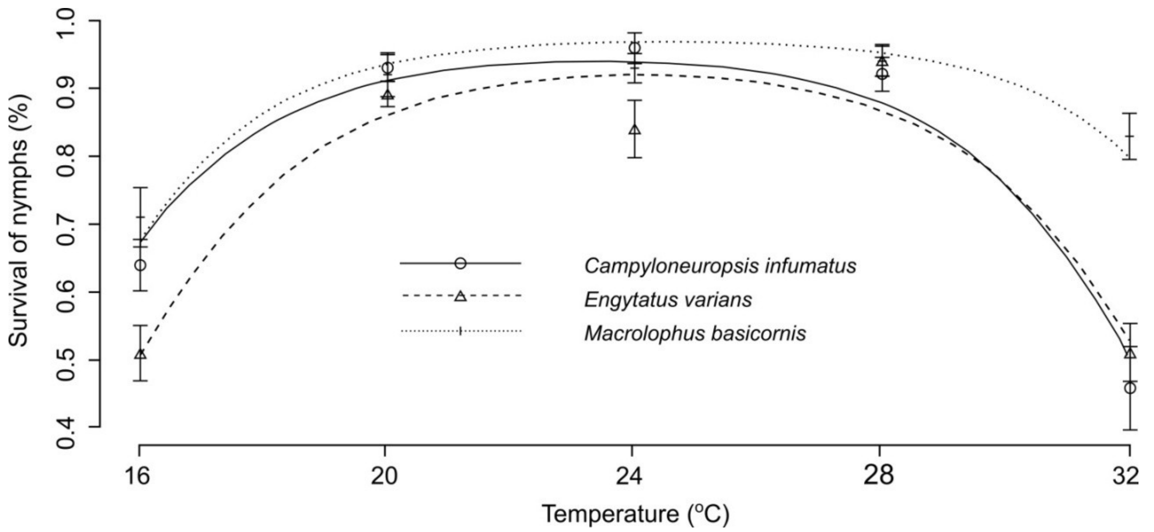

**Figure 5.** Fraction survival of nymphs (means ± SE) of three species of Miridae at five temperatures, RH 70 ± 10% and 12h photophase. *C. infumatus* $f(x)$=exp($-17.552295+1.735792x-0.037080x^2$)/[1+exp($-17.552295+1.73579\ 2x-0.037080x^22$)]; *E. varians* $f(x)$=exp($-17.552295-1.488780+1.73579\ 2x+0.049655x-0.037080x^2$)/[1+exp ($-17.552295-1.488780+1.735792x+0.049688x-0.037080x^2$)]; *M. basicornis* $f(x)$=exp($-17.552295-1.333229+1.735792x0.083811x-0.037080x^2$)/[1+exp($-17.552295-1.333229+1.735792x+0.083811x-0.037080x^2$).

**Table 3.** Regression equations and their correlation coefficients ($R^2$) used to calculate the lower developmental temperature thresholds (LTD) and thermal constants (K), for the different developmental stages of *C. infumatus*, *E. varians* and *M. basicornis*.

| Species | Stage | LTD (°C) | K(DD) | Regression equation | $R^2$(%) |
|---|---|---|---|---|---|
| *C. infumatus* | Egg | 7.94 | 156.25 | y=0.0064x-0.00508 | 0.99 |
| | Nymph | 9.81 | 232.56 | y=0.0043x-0.0422 | 0.98 |
| | Egg-adult | 9.35 | 384.62 | y=0.0026x-0.0243 | 0.99 |
| *E. varians* | Egg | 9.31 | 138.89 | y=0.0072x-0.0671 | 0.99 |
| | Nymph | 9.21 | 243.90 | y=0.0041x-0.0378 | 0.98 |
| | Egg-adult | 9.42 | 384.62 | y=0.0026x-0.0245 | 0.99 |
| *M. basicornis* | Egg | 8.65 | 166.67 | y=0.006x-0.0519 | 0.99 |
| | Nymph | 7.40 | 321.50 | y=0.0032x-0.0237 | 0.99 |
| | Egg-adult | 7.88 | 476.19 | y=0.0021x-0.0165 | 0.99 |

values of the lower developmental temperatures (LTD) of the development stages were different for all three mirid species (table 3). The values of LTD were lowest for the species *M. basicornis*. The thermal constants (K) for egg-adult development of *C. infumatus* and *E. varians* were similar and lower than that of *M. basicornis* (table 3). The thermal constant for the nymphal stage of *M. basicornis* was higher than that of *C. infumatus* and *E. varians* (table 3).

### Nymphal instar characteristics and weight of 4th and 5th nymphs

Newly emerged 1st instar nymphs of *C. infumatus* and *M. basicornis* were light green, whereas nymphs of *E. varians* were yellow. Nymphs of all three mirid species have brown-red eyes. First instar nymphs are of similar size (0.221 mm, measured by the largest width of the cephalic capsule between the outer margins of the compound eyes). The 2nd, 3rd, 4th and 5th nymphal instars of *C. infumatus* and *M. basicornis* were similar in cephalic capsule size (0.284, 0.292, 0.420 and 0.435 mm, respectively), while the respective instars of *E. varians* were slightly bigger (0.300, 0.376, 0.458 and 0.525 mm).

Differences in weight were found for the 5th nymphal instars of each mirid species reared at the five temperatures, but no clear trends were discovered, except that weight was generally lowest at the highest temperature (table 4). *C. infumatus* showed the lowest weights for the 4th ($F_{4;95}$ = 32.2310, $P < 0.0001$) and 5th nymphal instars ($F_{4;95}$ = 3.8040, $P < 0.0065$), at 32 °C. *E. varians* showed the lowest weight of the 4th (Dunn = 35.4221, df = 4, $P < 0.0001$) and 5th nymphal instars at 16 °C (Dunn = 63.1319, df = 4, $P < 0.0001$). *M. basicornis* showed lowest weight of the 4th (Dunn = 63.7721, df = 4, $P < 0.0001$) and 5th ($F_{4;95}$ = 43.5140, $P < 0.0001$) nymphal instars at 16 °C and 32 °C (table 4).

### Sex ratio

Temperature did not significantly affect sex ratios of the three mirid species. *E. varians* and *M. basicornis* exhibited a sex ratio which did not significantly differ from 1:1 at the five temperatures, whereas the sex ratio of *C. infumatus* was significantly female biased at all temperatures (table 5).

**Table 4.** Weight (mg) of 4th and 5th nymphal instars (means ± SE) of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase, n = 20 individuals.

| T °C | Species | Weight of nymphal instar (mg)<br>4th | 5th |
|---|---|---|---|
| 16 | *C. infumatus* | 0.970 ± 0.012A | 1.375 ± 0.031A |
| 20 | | 0.810 ± 0.026B | 1.210 ± 0.041B |
| 24 | | 0.815 ± 0.024B | 1.285 ± 0.024AB |
| 28 | | 0.700 ± 0.251C | 1.300 ± 0.034AB |
| 32 | | 0.610 ± 0.028C | 1.240 ± 0.027B |
| 16 | *E. varians* | 0.870 ± 0.026c | 1.370 ± 0.029d |
| 20 | | 0.845 ± 0.023c | 1.715 ± 0.033bc |
| 24 | | 1.000 ± 0.029ab | 1.970 ± 0.032a |
| 28 | | 0.895 ± 0.026bc | 1.885 ± 0.043ab |
| 32 | | 1.055 ± 0.019a | 1.620 ± 0.042c |
| 16 | *M. basicornis* | 0.750 ± 0.019b | 1.075 ± 0.032C |
| 20 | | 0.920 ± 0.015a | 1.380 ± 0.036B |
| 24 | | 0.810 ± 0.216b | 1.580 ± 0.031A |
| 28 | | 0.775 ± 0.012b | 1.185 ± 0.024C |
| 32 | | 0.565 ± 0.024c | 1.130 ± 0.031C |

Means followed by the same capital (Tukey test) or small (Dunn test) letters in columns are not significantly different when comparing each species among temperatures ($p < 0.05$). The Tukey test was applied when the data showed a normal distribution, the Dunn test was applied on data with a non-normal distribution.

**Table 5.** Sex ratio expressed as a proportion of females [SR = ♀ / (♀ + ♂)] of *C. infumatus*, *E. varians* and *M. basicornis* at five temperatures, RH 70 ± 10% and 12h photophase.

| T °C | Species | Sex ratio | $\chi^2$ |
|---|---|---|---|
| 16 | *C. infumatus* | 0.67 (43/21) | 7.5625* |
| 20 | | 0.87 (81/12) | 51.1936** |
| 24 | | 0.86 (83/13) | 51.0417** |
| 28 | | 0.84 (78/14) | 44.5217** |
| 32 | | 0.73 (34/12) | 10.5217** |
| 16 | *E. varians* | 0.63 (33/19) | 3.7692ns |
| 20 | | 0.41 (37/52) | 2.5281ns |
| 24 | | 0.55 (47/37) | 1.1905ns |
| 28 | | 0.50 (47/47) | 0.0000ns |
| 32 | | 0.52 (27/24) | 0.1765ns |
| 16 | *M. basicornis* | 0.54 (39/32) | 0.6901ns |
| 20 | | 0.47 (44/48) | 0.1739ns |
| 24 | | 0.59 (55/38) | 3.1075ns |
| 28 | | 0.52 (49/45) | 0.1702ns |
| 32 | | 0.56 (47/36) | 1.4578ns |

Significant difference by $\chi^2$ test at *5% ($\chi^2$ tabulated = 3.841, df = 1) and **1% ($\chi^2$ tabulated = 5.9915, df = 1); ns = not significant by $\chi^2$ test.

## Discussion

The performance of organisms depends greatly - but not only - on temperature, with performance generally being maximized within a rather narrow thermal range (Angilletta, 2009). Temperature affected most variables during immature development of the three mirid species, and temperature effects are also reflected in their lower threshold temperatures and thermal constants. *C. infumatus* and *E. varians* nymphs were heavier, and showed higher survival at intermediate temperatures. *M. basicornis* nymphs showed a high survival at all temperatures, but, like the other two mirids, was lighter at the lowest and highest temperature. *E. varians* appeared least adapted to the lowest temperature, indicated by low survival and weight. Only sex ratio was not affected by temperature. Temperature extremes of 16 °C and 32 °C had in most cases a negative effect on development and survival of eggs and nymphs. The negative effects at the lowest temperature are in agreement with findings of Roy *et al.* (2002), who concluded that exposure of insects to low temperature affects all of their development stages, with a reduction in the metabolic rate and, consequently, with slower development. However, all three species were still developing and showed immature survival at the lowest (> 42.5%) and highest (> 33%) temperatures, so they remain active at the optimal Brazilian temperature regime for tomato production (Naika *et al.*, 2006).

Though we also found negative effects at the highest temperature, egg survival values at 32 °C for *C. infumatus*, *E. varians* and *M. basicornis* were still higher than those found for *M. pygmaeus* at 30 °C (Perdikis and Lykouressis, 2002; 2004). Host plant effects - we used tobacco, while Perdikis and Lykouressis (2004) used tomato and eggplant plants as oviposition substrates - may partly explain the difference in egg survival. Another difference between their and our experiment is that we offered prey to the egg laying adults, while Perdikis and Lykouressis (2004) did not. Many species of mirids show endophytic oviposition, and their eggs, which are embedded in the leaf, are particularly vulnerable to desiccation (Wheeler, 2001). Cocco *et al.* (2008) mentioned that tobacco has a high cell turgidity and Constant *et al.* (1996) observed egg-hatching percentages of 81.6% on tobacco, while they found only 43.4% egg-hatching on geranium, *Pelargonium peltatum* L. (Geraniaceae), as oviposition substrates for

*M. pygmaeus*. Our results in combination with the above mentioned literature data suggest that tobacco provides a good oviposition substrate for mirids, whereas tomato, sweet pepper and gerbera might be less suitable.

The three mirid species showed five instar stages independent of the temperature to which they were exposed. Previous studies with other mirids, *M. pygmaeus* (Perdikis and Lykouressis, 2002) and *N. tenuis* (Urbaneja *et al*., 2005), also showed five instar stages when exposed to different temperatures. Exposure of the three mirids to 16 °C led to increased nymphal and egg-adult development time, probably due to a slower metabolism at low temperatures. Sinclair *et al*. (2003) reported that metabolic activity as well as the amount of food intake of insects decreases at low temperatures, while energy expenditure increases. The development times observed for *C. infumatus* and *M. basicornis* at 32 °C were similar to values reported for the mirid *Pilophorus typicus* (Distant) at 30 °C (Nishikawa *et al*., 2010). Temperature effects on nymphal survival were similar to those on development: survival was highest at intermediate temperatures, and was lower at the extremes, though survival of *M. basicornis* was less affected than that of the other two species at the extremes. Sanchez *et al*. (2009) reported highest survival for *N. tenuis* at 25 °C and lowest at 15 °C and 35 °C, while no nymphs survived at 40 °C. Nishikawa *et al*. (2010) observed lower survival rates of *P. typicus* at lower (17.5 °C) and higher (30 °C) temperatures.

Wheeler (2001) stated that latitudinal and altitudinal effects on the development of mirids species are evident. The lower temperature thresholds and thermal constants we found for three mirid species were quite different, despite the fact that they were collected at the same location, and were exposed to the same food and rearing conditions. Nevertheless, these three mirid species have a wide geographical distribution over the America's, and may, thus, have regional populations with different temperature thresholds and thermal constants like we found. *M. basicornis* starts nymphal development at a lower temperature threshold and needs more degree days than *C. infumatus* and *E. varians.* The values of the lower temperature thresholds we found were lower than those recorded for the mirid *N. tenuis* (Sanchez *et al*., 2009; Hughes *et al*., 2010; Pazyuk *et al*., 2014), indicating that the Neotropical species we studied start performing at lower temperature conditions than the mirids successfully used in biocontrol in Europe.

Although significant differences in weight were found for each mirid species reared at different temperatures, no clear trends were found, with the exception of the highest temperature where weight was generally lower than at the other temperatures. This is in agreement with Wheeler (2001), who reported that, in general, more rapid development at higher temperatures is associated with reduced weight and survivorship.

Temperature did not affect the sex ratio of the three mirids. Lauge (1985) stated that unfavourable conditions for nymphal development, such as extreme temperatures might affect sex ratios, resulting in fewer females, but we did not observe such an effect. Jervis and Copland (1996) reported that diploid species, such as hemipterans, show 1:1 sex ratios with little variation. This is what we found for *E. varians* and *M. basicornis.* Similar 1:1 ratios were found for the mirids *M. pygmaeus* (Perdikis and Lykouressis, 2004), *N. tenuis* (Sanchez *et al*., 2009) and *Tupiocoris cucurbitaceus* (Spinola) (López *et al*., 2012). However, the sex ratio of *C. infumatus* was strongly female biased, and as yet, we have no explanation for this finding.

In order to be efficient, natural enemies should be able to disperse, develop, reproduce and attack the pest under the weather conditions in which they are to be used (van Lenteren, 2010). The pest *T. absoluta* is active within a temperature range of 19.7 °C to 27.1 °C, with the shortest development time at 27.1 °C (23.8 days) (Barrientos *et al*., 1998). It shows a high egg, larval and pupal survival, and the shortest larval development time at 25 °C. Its lower temperature threshold is 8.14 °C and its thermal constant is 453.60 DD (Barrientos *et al*., 1998). All these values are in the range of those that we found for the three mirid predators. Thus, the pest optimally develops within the same temperature range as the immature stages of the predatory mirids. The overlap in performance at a range of temperatures is illustrated in figure 4, where development times of the pest and the mirids are given. Climate matching between pest and predators is a positive finding, as the predators can establish themselves and develop in crops where the prey is present and active.

Climate matching of pest and predator, together with a high predation capacity of the nymphs and adults of the three mirids (Bueno *et al*., 2013a; van Lenteren *et al*., 2016) may make these mirids potential candidates for biological control of *T. absoluta.* Based on climate matching data alone, *M. basicornis* might be the best candidate for biological control of this pest, as it has the lowest temperature threshold and is performing best at intermediate and extreme temperatures, but we do realize, of course, that a number of other predator characteristics will determine its eventual success in biocontrol of *T. absoluta*. We have recently published on population development of the three mirids on tomato with *T. absoluta* as prey (Silva *et al*., 2016), with *E. kuehniella* as prey (Bueno *et al*., 2018), on predation of prey by all nymphal stages and during the whole adult stage (van Lenteren *et al*., 2017; 2018c), about the effect of plant and fruit feeding by these mirids (Silva *et al*., 2017a, van Lenteren *et al*., 2018b), on the role of chemical communication in the mirid-pest-plant system (Silva *et al*., 2017b; 2018), as well as on the capacity to reduce pest populations in an experimental greenhouse setting (van Lenteren *et al*., 2018b). All current information of these three Neotropical mirids is now under assessment and will be used for selection of the most promising predator for control of *T. absoluta* and other pests on tomato in Brazil.

An additional conclusion of this study is that tobacco is a good host plant and *E. kuehniella* eggs are suitable factitious prey for the three mirids. Mass rearing of the mirids under laboratory conditions is much easier on tobacco than tomato, and *E. kuehniella* eggs are easier to obtain than *T. absoluta* eggs.

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## References

Agrianual, 2016.- *Anuário estatístico da Agricultura Brasileira*.- FNP, São Paulo, Brazil.

Albajes R., Alomar O., 2008.- Facultative Predators, pp.1400-1405. In: *Encyclopedia of entomology* (Capinera J. L., Ed.).- Springer, Dordrecht, The Nehterlands.

Angilletta M. J., 2009.- *Thermal adaptation: a theoretical and empirical synthesis*.- Oxford University Press, Oxford, UK.

Aubry O., Cormier D., Chouinard G., Lucas E., 2015.- Influence of plant, animal and mixed resources on development of the zoophytophagous plant bug *Campylomma verbasci* (Hemiptera: Miridae).- *Biocontrol Science and Technology*, 25: 1426-1442.

Barrientos Z. R., Apablaza H. J., Norera S. A., 1998.- Temperatura base y constante térmica de desarrollo de la polilla del tomate, *Tuta absoluta* (Lepidoptera: Gelechiidae).- *Ciencia e Investigación Agraria*, 25: 133-137.

Bergant K., Trdan S., 2006.- How reliable are thermal constants for insect development when estimated from laboratory experiments.- *Entomologia Experimentalis et Applicata*, 120: 251-256.

Bueno V. H. P., van Lenteren J. C., 2012.- Predatory bugs (Heteroptera), pp. 539-569. In: *Insect bioecology and nutrition for integrated pest management* (Panizzi A. R., Parra J. R. P., Eds).- CRC Press, Boca Raton, Florida, USA.

Bueno V. H. P., Montes F. C., Calixto A. M., Lins Jr J. C., van Lenteren J. C., 2012.- Can recently found Brazilian hemipteran predatory bugs control *Tuta absoluta*?- *IOBC/wprs Bulletin*, 80: 63-67.

Bueno V. H. P., van Lenteren J. C., Lins Jr J. C., Calixto A. M., Montes F. C., Silva D. B., Santiago L. D., Pérez L. M., 2013a.- New records of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) predation by Brazilian Hemipteran predatory bugs.- *Journal of Applied Entomology*, 137: 29-34.

Bueno V. H. P., van Lenteren J. C., Lins Jr J. C., Montes F. C., Calixto A. M., Silva D. B., 2013b.- Biological control of *Tuta absoluta*: sampling and evaluation of new hemipteran predators found in Brazil, pp. 269-272. In: *Proceedings of the 4th international symposium on biological control of arthropods* (Mason P. G., Gillespie D. R., Vicent C., Eds), Agriculture and Agri-Food, Canada.

Bueno V. H. P., Calixto A. M., Montes F. C., van Lenteren J. C., 2018.- Population growth parameters of three Neotropical mirid predators (Hemiptera: Miridae) at five temperatures on tobacco with *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) eggs as food.- *Israel Journal of Entomology*, 48 (2): 1-22.

Calvo J. F., Lorente M. J., Stansly P. A., Belda J. E., 2012.- Preplant release of *Nesidiocoris tenuis* and supplementary tactics for control of *Tuta absoluta* and *Bemisa tabaci* in greenhouse tomato.- *Entomologia Experimentalis et Applicata*, 143: 111-119.

Campbell A., Frazer B. D., Gilbert N., Gutierrez A. P., Mackauer M., 1974.- Temperature requirements of some aphids and their parasites.- *Journal of Applied Ecology*, 11: 431-438.

Clarke A., 2017.- *Principles of thermal ecology*.- Oxford University Press, Oxford, UK.

Cocco C., Santos O. S., Santos P. M., Carvalho M. P., Lopes S. J., 2008.- Irrigation systems and containers for the production of tobacco runners in hydroponics.- *Revista Brasileira de Agrociências*, 14: 27-37.

Constant B., Grenier S., Bonnot G., 1996.- Artificial substrate for egg laying and embryonic development by the predatory bug *Macrolophus caliginosus* (Heteroptera: Miridae).- *Biological Control*, 7: 140-147.

Guedes R. N. C., Picanço M. C., 2012.- The tomato borer *Tuta absoluta* in South America: pest status, management and insecticide resistance.- *EPPO Bulletin*, 42: 211-216.

Gujarati D. N., 2004.- *Basic ecomometrics*, 4th edition.- McGraw Hill, New York, USA.

Horn D. J., 1998.- Temperature synergism in integrated pest management, pp. 125-139. In: *Temperature sensitivity in insects and application in integrated pest management* (Hallman G. J., Delinger D. L. Eds).- Westview Press, Boulder, CO, USA.

Hughes G. E., Alford L., Sterk G., Bale J. S., 2010.- Thermal activity thresholds of the predatory mirid *Nesidiocoris tenuis*: implications for its efficacy as a biological control agent.- *BioControl*, 55: 493-501.

Jaworski C. C., Bomprd A., Genies L., Amiens-Desneux E., Desneux N., 2013.- Preference and prey switching in a generalist predator attacking local and invasive alien pests.- *Plos ONE*, 8 (12): e82231.

Jervis M. A., Copland M. J. W., 1996.- The life cycle, pp. 63-160. In: *Insect natural enemies: practical approaches to their study and evaluation* (Jervis M.A., Kidd N. Eds).- Chapman and Hall, London, UK.

Lauge G., 1985.- Sex determination: genetic and epigenetic factors, pp. 295-318. In: *Comprehensive insect physiology, biochemistry and pharmacology - embryogenesis and reproduction*, 1st edition (Kerkut G. A., Gilbert L. I. Eds).- Pergamon, Oxford, UK.

Lee Y., Nelder J. A., Pawitan Y., 2006.- *Generalized linear models with random effects*.- Chapman and Hall, New York, USA.

López S. N., Rojas F. A., Velásquez V. V., Cagnotti C., 2012.- Biology of *Tupiocoris cucurbitaceus* (Hemiptera: Miridae), a predator of the greenhouse whitefly *Trialeurodes vaporariorum* (Hemiptera: Aleyrodidae) in tomato crops in Argentina.- *Biocontrol Science and Technology*, 22: 1107-1117.

Martínez A. M., Baena M., Figueroa J. I., Del Estal P., Medina M., Guzman-Lara E, Pineda S., 2014.- Primer registro de *Engytatus varians* (Distant) (Hemiptera: Heteroptera: Miridae) em México y su depredación sobre *Bactericera cockerelli* (Sulc) (Hemiptera: Triozidae): uma revisíon de su distribución y hábitos.- *Acta Zoológica Mexicana*, 30: 617- 624.

McCullagh P., Nelder J. A., 1989.- *Generalized linear models*, 2nd edition.- Chapman and Hall, London, UK.

Mollá O., Biondi A., Alonso-Valiente M., Urbaneja A., 2014.- A comparative life history study of two mirid bugs preying on *Tuta absoluta* and *Ephestia kuehniella* eggs on tomato crops: implications for biological control.- *BioControl*, 59: 175-183.

Naika S., van Lidt de Jeude J., de Goffau M., Hilm M., van Dam B., 2006.- *A cultura do tomate: produção, processamento e comercialização*, Agrodok 17.- Fundação Agromisa e CTA, Wageningen, The Netherlands.

Nelder J. A., Wedderburn R. W. M., 1972.- Generalized Linear Models.- *Journal of the Royal Statistical Society A*, 135: 370-384.

Nishikawa H., Shimada T., Nakahira K., Arakawa R., 2010.- Thermal effect on the development and reproduction of an indigenous mirid bug, *Pilophorus typicus* Distant (Heteroptera: Miridae), a potential biological control agent in Japan.- *Applied Entomology and Zoology*, 45: 313-318.

Pazyuk I. M., Musolin D. L., Reznik S. Y., 2014.- Geographic variation in thermal and photoperiodic effects on development of zoophytophagous plant bug *Nesidiocoris tenuis*.- *Journal of Applied Entomology*, 138: 36-44.

Perdikis D. C., Lykouressis D. P., 2002.- Life table and biological characteristics of *Macrolophus pygmaeus* when feeding on *Myzus persicae* and *Trialeurodes vaporariorum*.- *Entomologia Experimentalis et Applicata*, 102: 261-272.

Perdikis D. C., Lykouressis D. P., 2004.- *Macrolophus pygmaeus* (Hemiptera: Miridae) population parameters and biological characteristics when feeding on eggplant and tomato without prey.- *Journal of Economic Entomology*, 97: 1291-1298.

Pérez-Hedo M, Urbaneja A, 2016.- The zoophytophagous predator *Nesidiocoris tenuis*: a successful but controversial biocontrol agent in tomato crops, pp. 121-138. In: *Advances in insect control and resistance management* (Horowitz A. R., Ishaaya I., Eds).- Springer, Dordrecht, The Netherlands.

Pérez-Hedo M., Urbaneja-Bernat P., Jaques J. A., Flors V., Urbaneja A., 2015.- Defensive plant responses induced by *Nesidiocoris tenuis* (Hemiptera: Miridae) on tomato plants.- *Journal of Pest Science*, 88: 543-554.

R Development Core Team, 2015.- *R: a language and environment for statistical computing*.- R Foundation for Statistical Computing, Vienna, Austria.

Roy M., Brodeur J., Cloutier C., 2002.- Relationship between temperature and development fate of *Sthetorus punctillum* (Coleoptera: Coccinellidae) and its prey *Tetranychus mcdalieli* (Acarina: Tetranichidae).- *Environmental Entomology*, 31: 177-186.

Sanchez J. A., Lacasa A., Arno J., Castane C., Alomar O., 2009.- Life history parameters for *Nesidiocoris tenuis* (Reuter) (Het., Miridae) under different temperature regimes.- *Journal of Applied Entomology*, 133: 125-132.

Silva G. A, Picanço M. C., Bacci L., Crespo A. L. B., Rosado J. F., Guedes R. N. C., 2011.- Control failure likelihood and spatial dependence of insecticide resistance in the tomato pinworm, *Tuta absoluta*.- *Pest Management Science*, 67: 913-920.

Silva D. B., Bueno V. H. P., Montes F. C., van Lenteren J. C., 2016.- Population growth of three mirid predatory bugs feeding on eggs and larvae of *Tuta absoluta* on tomato.- *BioControl*, 61: 545-553.

Silva D. B., Bueno V. H. P., Calvo F. J, van Lenteren J. C., 2017a.- Do nymphs and adults of three Neotropical zoophytophagous mirid damage leaves and fruits of tomato?- *Bulletin of Entomological Research*, 107: 200-207.

Silva D. B., Weldegergis B., Van Loon J. J. A., Bueno V. H. P., 2017b.- Comparative analysis of herbivore-induced plant volatiles from tomato plants infested by either *Tuta absoluta* or *Bemisia tabaci*. - *Journal of Chemical Ecology* 43: 53-65.

Silva D. B., Bueno V. H. P., Van Loon J. J. A., Peñaflor M. F. G. V., Bento J. M. S., van Lenteren J. C., 2018.- Attraction of three mirid predators to tomato infested by both the tomato leaf mining moth *Tuta absoluta* and the whitefly *Bemisia tabaci*.- *Journal of Chemical Ecology*, 44: 29-39.

Sinclair B. J., Vernon P., Klok C. J., Chown S., 2003.- Insects at low temperatures: an ecological perspective.- *Trends in Ecology & Evolution*, 18: 257-262.

Siqueira H. A. A., Guedes R. N. C., Picanço M. C., 2000.- Insecticide resistance in populations of *Tuta absoluta* (Lepidoptera: Gelechiidae).- *Agricultural and Forest Entomology*, 2: 147-153.

Siqueira H. A. A., Guedes R. N. C., Fragoso D. B., Magalhães L. C., 2001.- Abamectin resistance and synergism in Brazilian populations of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae).- *International Journal of Pest Management*, 47: 247-251.

Thomazini A. P. B. W., Vendramim J. D., Brunherotto R., Lopes M. T. R., 2001.- Effect of *Lycopersicom* spp. genotypes on biology and oviposition of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae).- *Neotropical Entomology*, 30: 283-288.

Urbaneja A., Tapia G., Stansly P., 2005.- Influence of host plant and prey availability on developmental time and survivorship of *Nesidiocoris tenius* (Heteroptera: Miridae).- *Biocontrol Science and Technology*, 15: 513-518.

Vandekerkhove B., De Puysseleyr V., Bonte M., De Clercq P., 2011.- Fitness and predation potential of *Macrolophus pygmaeus* reared under artificial conditions.- *Insect Science*, 18: 682-688.

van den Bosch R., 1978.- *The pesticide conspiracy*.- University of California Press, Berkely, USA.

van Lenteren J. C., 2010.- *Ecology: cool science, but does it help*?- Wageningen University, Wageningen, The Netherlands.

van Lenteren J. C., 2012.- The state of commercial augmentative biological control: plenty of natural enemies, but a frustrating lack of uptake.- *BioControl*, 57: 1-20.

van Lenteren J. C., Hemerik L., Lins Jr J. C., Bueno V. H. P., 2016.- Functional responses of three Neotropical mirid predators to eggs of *Tuta absoluta* on tomato.- *Insects*, 7 (3): 34 doi:10.3390/insects70300347

van Lenteren J. C., Bueno V. H. P., Smit J., Soares M. A., Calixto A. M., Montes F. C., de Jong P., 2017.- Predation of *Tuta absoluta* eggs during the nymphal stages of three Neotropical mirid predators on tomato.- *Bulletin of Insectology*, 70 (1): 69-74.

van Lenteren J. C., Bolckmans K., Kohl J., Ravensberg W. J., Urbaneja A., 2018a.- Biological control using invertebrates and microorganisms: plenty of new opportunities.- *BioControl*, 63 (1): 39-59.

van Lenteren J. C., Bueno V. H. P., Calvo F. J., Calixto A. M., Montes F. C., 2018b.- Comparative effectiveness and injury to tomato plants of three Neotropical mirid predators of *Tuta absoluta* (Lepidoptera: Gelechiidae).- *Journal of Economic Entomology*, 111: in press (doi: 10.1093/jee/toy057).

van Lenteren J. C., Bueno V. H. P., Montes F. C., Hemerik L., De Jong P. W., 2018c.- Adult lifetime predation of *Tuta absoluta* eggs by three Neotropical mirid predators on tomato.- *Bulletin of Insectology*, 71 (2): in press.

Wheeler A. G., 2001.- *Biology of the plant bugs (Hemiptera: Miridae): pests, predators, opportunists*.- Cornell University Press, Ithaca and London.

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