Key words. Diptera, Tephritidae, Bactrocera dorsalis, Hymenoptera, Braconidae, Diachasmimorpha longicaudata, parasitoid, altering temperatures, mass-reared, activity of antioxidants

Abstract. Diachasmimorpha longicaudata is the most widely used endoparasitoid in biocontrol programmes against various species of tephritids and is continuously mass-reared under laboratory conditions at a constant temperature; however, little is known about how varying the temperature might affect the effectiveness of the mass rearing of immature D. longicaudata. This study aimed to determine the optimum temperature for the development of larvae D. longicaudata the larvae of its host, Bactrocera dorsalis. Third-instar B. dorsalis were exposed for 4 h to five pairs of parasitoids, and then they were kept at one of six temperatures ranging from 19°C to 34°C. The activities of three major antioxidant enzymes (CAT, POX, SOD) were individually measured 4, 24, 48 and 72 h after exposure to each of the six temperatures. The results were as follows: for CAT the highest CAT activities were identically at 25°C (intervals 0–4 h, 5–24 h) and at 31°C (interval 25–48 h, 49–72 h), respectively. Subsequently, the effects keeping the parasitized larvae at these different temperatures on the biological attributes of D. longicaudata reared under laboratory conditions were investigated. The percentage of their eggs that hatched, percentage of adults that emerged, pupal weight and longevity were significantly greater than that recorded for the control when the parasitized host larvae were kept at 28°C for 4 h, 25°C for 5 and 48 h and 31°C for 49 and 72 h.

1. INTRODUCTION

The oriental fruit fly Bactrocera dorsalis (Hendel) (Diptera: Tephritidae) is a generalist pest that feeds on more than 270 species of host plants in 46 different families of plants throughout tropical and subtropical Asia (Allwood & Chinajariyawong, 1999; Clarke et al., 2001; Vargas et al., 2007). This species is a devastating pest of carambola, peach, jujube, citrus, loquat, guava, lychee, longan and other tropical and subtropical crops, causing significant yield and financial losses in southern China (Ji et al., 2016). Thus, its control and management are of great socioeconomic significance. Great efforts were made over the last several decades to develop effective means of controlling B. dorsalis, however, in China, the control of this pest depends mainly on the use of chemical insecticides (Zhang et al., 2014). The use of wide-spectrum chemical pesticides results in many secondary problems, such as environmental pollution, decreases in the abundance of natural enemies, presence of chemical residues in foods and resistance of this pest to pesticides (Zhang et al., 2014; Cai et al., 2017).

Due to the ineffectiveness and undesirable consequences of using pesticides, parasitoid species, such as Fopius arisanus (Sonan), Psyttalia incisi (Silvestri), Fopius vandensorchis (Fullaway) and Diachasmimorpha longicaudata (Ashmead) (Hymenoptera: Braconidae), have been widely used to depress fruit fly populations in many regions (Vargas et al., 1993, 2012; Cai et al., 2017; Yang et al., 2018). Among the fruit fly parasitoids available for augmentative release in Fujian Province of China, D. longicaudata is the most reliable (Cai et al., 2018a).
Native to Southeast Asia, *D. longicaudata* is a koinobiont endoparasitoid that has been widely used in biological control programmes against tephritid fruit flies, including *B. dorsalis*, in southern China (Liu et al., 2012; Segura et al., 2016; Simmonds et al., 2016). *D. longicaudata* can become established in various environments and is amenable to mass rearing in the larvae of several species of the genus *Bactrocera* (Wharton & Gilstrap, 1983; Baranowski et al., 1993; Chinajariyawong et al., 2000). The role of *D. longicaudata* in biological pest control programmes has stimulated research into various aspects of its biology, such as conditions for mass rearing and release, molecular biology and physiology, in order to improve its use in agriculture pest management (Constanza et al., 2016; Costa et al., 2016; Ndlela et al., 2016; Simmonds et al., 2016; Julsirikul et al., 2017).

Evaluation of the optimum temperature for development of a parasitoid is an important step in its mass production under laboratory conditions and inductive release in the field and a basic requirement of any biological pest control programme (Meirelles et al., 2015). Temperature is one of the most important climatic factors influencing the distribution, abundance, immature development, adult emergence, fecundity, longevity and parasitism capability of egg parasitoids (Maceda et al., 2003; Liu et al., 2012). The effectiveness of biological control programmes can be limited by adverse climatic conditions (Firake & Khan, 2014). The effects of temperature on *D. longicaudata* developing in different hosts is well studied; for instance, Meirelles et al. (2013, 2015) report that rapid development of *D. longicaudata* in *Anastrepha fraterculus* (Wiedemann) and *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) is achieved at a constant temperature of 25°C. Liu et al. (2012) suggest that the most appropriate temperature range for the reproduction and development of *D. longicaudata* adults that developed as larvae in *B. dorsalis* is from 24 to 27°C. These previous studies investigated the effects of constant temperatures (Liu et al., 2012; Meirelles et al., 2013, 2015). However, parasitoids reared under constant laboratory conditions for a long period can upon release be adversely affected by different temperatures, which can influence their effectiveness as biological control agents (Firake & Khan, 2014).

To counter this, it is necessary to acclimatize the parasitoids to temperature fluctuations during rearing in the laboratory. The effects of varying the temperature on adults of *D. longicaudata* are well studied (Liu et al., 2012; Meirelles et al., 2013, 2015), however, its effect on immature stages is often ignored. The basic daily thermal requirements of larvae of *D. longicaudata* developing within host larvae were evaluated based on the effects of varying the temperature on the levels of three major antioxidant enzymes in parasitized larvae of *B. dorsalis* at different stages in their development.

In healthy animals, a balance exists between the production of reactive oxygen species (ROS) and antioxidant processes. However, various types of stress, e.g., thermal stress and parasitization, can disrupt the antioxidant system and induce oxidative stress (Carton et al., 2008; Lopez-Martinez et al., 2008). The overexpression of ROS in the form of the superoxide (O$_2^-$), hydroxyl radicals (OH·), hydrogen peroxide (H$_2$O$_2$) and singlet oxygen (O$_2^*$) can result in an increase in protein oxidation, DNA damage and lipid peroxidation (LPO), which may lead to apoptosis due to enzyme inactivity and membrane damage (Livingstone, 2001; Yang et al., 2010). To maintain homeostasis and prevent oxidative damage, insects have evolved a complex enzymatic and non-enzymatic defence system to mitigate oxidative stress. The main antioxidant enzymes in insects are superoxide dismutase (SOD), catalase (CAT) and peroxidases (POX) (Dubovskiy et al., 2008; Ali et al., 2017). SOD breaks down superoxide radicals into oxygen and H$_2$O$_2$, whereas both CAT and POX convert H$_2$O$_2$ into oxygen and water (Yang et al., 2010). A significant increase in the activity of the antioxidant enzymes, CAT, POX and SOD is a sign of oxidative stress and an indicator of the ability of insects to combat oxidative stress by cooperatively eliminating the oxidative stress resulting from the high concentrations of ROS in cells (Jia et al., 2011; Cai et al., 2018b).

Previous studies have indicated that at a constant temperature the activity of POX in larvae of *B. dorsalis* is significantly increased by infection with *P. incisii* (Liang et al., 2007). CAT, POX and SOD, together, have an important role in preventing the larvae of *B. dorsalis* suffering from oxidative damage induced by parasitization by *D. longicaudata* (Wang et al., 2013). The effects of different temperatures on the adults of *B. dorsalis* indicate it is likely that these three main antioxidant enzymes have an important role in reducing oxidative stress induced by thermal stress (Jia et al., 2011). Nevertheless, there are few studies on the effects of different temperatures on parasitized agriculture pests. In this study, we determined the effects of temperature on the three main antioxidant enzymes in parasitized larvae of *B. dorsalis* after different periods of time in order to determine the most appropriate thermal strategy for rearing parasitized hosts and their parasitoid progeny. Subsequently, the effects of different temperatures on the biological attributes of *D. longicaudata* reared under laboratory conditions were determined. This study aimed at increasing the efficiency of the mass-rearing of *D. longicaudata* and the performance of this parasitoid in the field by varying the temperature at which the immature stages of this parasitoid were reared.

### 2. METHODS

#### 2.1. Insects

Colonies of *B. dorsalis* and *D. longicaudata* were established initially by collecting rotten citrus fruit infested with *B. dorsalis* from an orchard in Zhejiang city, Fujian, China, and then reared in the Beneficial Insects Institute of Fujian Agriculture and Forestry University (BII, FAFU), Fuzhou, Fujian, China, in accordance with the procedures reported by Spencer & Fujita (1997) and Shao et al. (2008). The *B. dorsalis* larvae were cultured on an artificial diet based on a mixture of torula yeast, wheat bran, sugar, sodium benzoate, nipagin and water in fixed proportions. The adult flies were kept in a cage (30 × 30 × 30 cm) under...
controlled conditions at 25 ± 1°C, 65 ± 5% relative humidity and a photoperiod of 12L : 12D, and provided with water and a mixture of torula yeast and sugar. The *D. longicaudata* were reared in third-instar larvae of *B. dorsalis*, and the parasitoid adults were fed a 10% honey solution in a Hawaii-type cage (Wong & Ramanadan, 1992) (30 × 30 × 30 cm) under the same laboratory conditions as above.

### 2.2 Experiments

#### 2.2.1.1 Parasitization by *D. longicaudata*

When the larvae of *B. dorsalis* reached the third stage (five days after collecting the eggs), they were transferred from the artificial diet to an oviposition unit consisting of a Petri dish with a mesh top (height: 0.5 cm; diameter: 6 cm, Aluja et al., 2009). Each oviposition unit contained a small amount of larval diet and a random sample of 100 larvae. Then, the oviposition units were placed in cages and exposed to five pairs of 7-day-old virgin parasitoids (female : male = 1 : 1) for 4 h at a constant temperature of 25°C. To reduce superparasitism by *D. longicaudata*, the ratio of female parasitoids to fruit flies was 20 to 1 (Lin et al., 2006). After 4 h of exposure, the larvae were placed on larval diet on a plate (4 × 10 × 20 cm). Based on the reported optimum temperature for adults of *D. longicaudata* (Liu et al., 2012), six temperatures ranging from 19°C to 34°C were selected for rearing parasitized larvae of *B. dorsalis*. Then each of the plates was placed in one of six climatic cabinets that were each set at either 19°C, 22°C, 25°C, 28°C, 31°C or 34°C (PRX-80A, Shanghai Joe Yue Electronics Co. Ltd., Shanghai, China), respectively. A constant humidity (70%) and photoperiod (12L : 12D) throughout the evaluation.

#### 2.2.1.2 Sample preparation

Four hours after parasitization, five live parasitized *B. dorsalis* larvae from each treatment were selected at random and washed more than three times using distilled water. The parasitoid larvae were quickly removed from the host larvae using a dissecting needle under a microscope. To eliminate potential confounding effects of superparasitism, host larvae that contained more than one parasitoid larva were excluded from the assays. After dissection, the remains of the bodies of the *B. dorsalis* larvae were immediately frozen in liquid nitrogen and stored at −80°C until analyzed, and the remaining *B. dorsalis* larvae were returned to their respective climatic cabinets to continue their development. This was repeated 24, 48 and 72 h after parasitization.

#### 2.2.1.3 Protein extraction

Total protein was extracted from samples using commercially available kits from Shanghai Lanji Bioengineering Institute (Shanghai, China) following the manufacturer’s instructions. Prepared fruit fly samples were first homogenized in 2 mL of ice-cold 0.05 M pH 7.0 phosphate buffer (PB). The crude homogenates were centrifuged at 8000 r/min for 10 min at 4°C, and the supernatant was used for further analysis. Protein concentrations were evaluated in accordance with the Bradford (1976) method using serum albumin as the standard.

#### 2.2.1.4 The antioxidant enzyme activity assay

The activities of CAT, POX and SOD were evaluated based on the protocols of the commercially available assay kits used (Shanghai Lanji Bioengineering Institute, Shanghai, China). Absorbance was read using a microplate spectrophotometer (UV2900, Sunny Optical Technology Co., Ltd., China).

CAT activity was calculated by determining the decrease in absorbance at 230 nm due to *H₂O₂* decomposition (Luck, 1971). One unit of CAT activity was defined as the amount of *H₂O₂* decomposed per second per gram of protein. CAT activity was expressed as U g⁻¹ protein.

The POX activity was calculated spectrophotometrically at 470 nm based on the catalysis of oxidation in the presence of *H₂O₂* (Reddy et al., 1985). One unit of POX activity was described as the amount that catalyses 1 μg of substrate in one minute per milligram of protein. POX activity was expressed as U g⁻¹ protein.

The SOD activity was calculated at 560 nm using xanthine and xanthine oxidase systems (Marklund & Marklund, 1974). One unit of SOD activity was defined as the amount of enzyme required to decrease the xanthine and xanthine oxidase system reaction in 1 mg/mL protein extract by 50%. SOD activity was expressed as U mg⁻¹ protein.

#### 2.2.2 Experiment II: Effects of different temperatures on biological attributes of *D. longicaudata*

When the adults of *B. dorsalis* in the laboratory colony were sexually mature (10 days after emergence), their eggs were collected using a bottle containing orange juice and then uniformly distributed on the larval diet and reared under constant conditions (temperature, 25°C; humidity, 70%; photoperiod, 12L : 12D) until parasitized larvae of *B. dorsalis* were transferred from the artificial diet to an oviposition unit with a mesh top; each unit contained a small amount of larval diet (height: 0.5 cm; diameter: 6 cm, Aluja et al., 2009). Then, five of these units were placed in a cage and exposed to twenty-five pairs of 7-day-old parasitoids (female : male = 1 : 1) for 4 h under controlled conditions as described above.

After 4 h of exposure, all the oviposition units were placed in climatic chambers kept at different temperatures (see Table 1). Parasitoids reared at a constant temperature of 25°C served as the untreated control. The exposed larvae were transferred to the artificial diet and kept in a box (4 × 10 × 20 cm) at a constant humidity (70%) and photoperiod (12L : 12D) throughout the evaluation.

The temperature treatments, a random sample of 50 larvae were dissected to check the percentage of the parasitoid eggs that hatched, which was calculated as the number of eggs that hatched divided by the number of parasitized larvae × 100. This experiment was replicated three times.

Then, the incubation boxes with the remaining larvae were transferred from the climatic chambers into larger boxes with mesh tops (10 × 30 × 50 cm) and kept under constant conditions of temperature, 25°C humidity 70% and photoperiod, 12L : 12D. Moist sand was spread on the bottoms of the large boxes for the larvae to pupate in. On the sixth day after pupation, random samples of 300 blackening pupae from each treatment were washed and dried at room temperature and then weighed. The weighed pupae were returned to covered Petri dishes for emergence. After

<table>
<thead>
<tr>
<th>Interval of time interval (h)</th>
<th>CAT</th>
<th>POX</th>
<th>SOD</th>
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<tr>
<td>4</td>
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The values in the table are degrees Celsius (°C).
emergence, the number of adult flies and parasitoids were calculated for each treatment to estimate the percentage emergence and sex ratio. There were three replicates of all treatments. To record longevity, 50 male and 50 female parasitoids from each treatment were placed in a Hawaii-type cage with water and an adult diet consisting of 10% honey. Mortality was evaluated daily until all parasitoids had died.

2.3 Statistical analysis

All statistical procedures were conducted using SPSS 17.0 (SPSS Inc., Chicago, Illinois, USA) and WPS Office 2017 software (Kingsoft Corporation Limited, Beijing, China). The data on enzyme activity recorded in different treatments were subjected to a two-way analysis of variance (ANOVA) (temperature and period) using the general linear model procedure in SPSS 17.0. Average differences were subjected to the least significant difference (LSD) test and considered significant if differences were subjected to the least significant difference (LSD) test and considered significant if the F-values were significant (P < 0.05). The percentage of parasitoid eggs that hatched (%) and sex ratio were analyzed using one-way ANOVA and Tukey’s multiple range test with differences considered significant at P < 0.05. All percentages were square root transformed to improve normality and homoscedasticity (Zar, 1984), but untransformed means are presented in the figures. Longevity was analyzed using the log-rank test.

3. RESULTS

3.1. Experiment I

The mortality of oriental fruit fly larvae parasitized by D. longicaudata reared at the different temperatures ranged from 13.6% to 21.6%. Dead fruit fly larvae were excluded from the evaluation.

The statistical analysis revealed that the activity of CAT in parasitized larvae of B. dorsalis was significantly affected by temperature (F = 4.25, df = 5, 24; P < 0.01) and the time for which they were exposed to the temperature (F = 2.75, df = 3, 24; P < 0.01). The interaction between temperature and time interval was also significant (F = 4.59, df = 15, 24; P < 0.01). After 4 h, the activity of CAT in B. dorsalis was not significantly different from those recorded after the other intervals, whereas the activity after this interval of time was lower than after all the other periods and peaked at 28°C. The activity of CAT recorded after 5 and 24 h, and 25 and 48 h exhibited similar trends with increase in temperature and both peaked at 25°C. After 48 h, the activity of CAT was highest at 31°C (Fig. 1).

The activity of POX in parasitized larvae of B. dorsalis was significantly affected by temperature (F = 42.14, df = 5, 24; P < 0.01) and time interval (F = 21.74, df = 3, 24; P < 0.01), and there was a significant interaction between temperature and time interval (F = 14.53, df = 15, 24; P < 0.01). There was a similar trend in the activity of POX with increase in temperature after 4 h and after 25 and 48 h. The highest values were recorded after 4 h at 25°C and after 25 and 48 h at 31°C. For the time intervals of 49 and 72 h, the activity of POX gradually increased with increase in temperature, peaking at 31°C, and was then less at 34°C. The activity of POX in B. dorsalis was significantly higher after 4 and 24 h than after other periods of time, except at 28°C, and peaked at 25°C (Fig. 2).

The activity of SOD in parasitized larvae of B. dorsalis was significantly enhanced by the different temperature treatments (F = 52.52, df = 5, 24; P < 0.01) and after the four time intervals (F = 34.45, df = 3, 24; P < 0.01). The interaction between temperature and time interval was significant (F = 25.73, df = 15, 24; P < 0.01). After 4 h, the activity of SOD in parasitized larvae of B. dorsalis was higher than after any of the other time intervals and peaked at 25°C. The trends in the activity of SOD in parasitized larvae of B. dorsalis after 5 and 24 h, 25 and 48 h, and 49 and 72 h were similar. The highest values were recorded after 5 and 24 h at 25°C, after 25 and 48 h at 31°C and after 49 and 72 h at 31°C (Fig. 3). In order to clarify the results of experiment I, the temperatures associated with the highest activities of the three main antioxidant enzymes in parasitized larvae of B. dorsalis after the different time intervals are listed in Table 2.

3.2. Experiment II

The percentage of eggs that hatched, pupal weight and percentage of the parasitoids that emerged in temperature
treatment 1 (after 4 h at 28°C, after 5 and 48 h at 25°C and after 49 and 72 h at 31°C) were significantly higher than those recorded in the other treatments, including the control (Percentage egg hatch: F = 0.56, df = 4, P < 0.05, Fig. 4; Pupal weight: F = 5.78, df = 4, P < 0.05, Fig. 5; Percentage parasitoid emergence: F = 9.15, df = 4, P < 0.05, Fig. 6). Significant increases were also recorded in the longevity of male and female parasitoids in temperature treatment 1 (after 4 h at 28°C, after 5 and 48 h at 25°C, after 49 and 72 h at 31°C) relative to that recorded in the other treatments (Male: χ^2 = 6.95, df = 4, P < 0.05; Female: χ^2 = 5.72, df = 4, P < 0.05, Fig. 8). However, the sex ratio of the parasitoid (female/male) was significantly lower in four of the temperature treatments (after 4 h at 28°C, 5 and 48 h at 25°C, 49 and 72 h at 31°C) and 2 treatments (after 48 h at 25°C, 49 and 72 h at 31°C) compared with the control treatment and temperature treatment 4 (F = 423.71, df = 4, P < 0.05, Fig. 7).

4. DISCUSSION

The ecological compatibility of a biological control agent is an important aspect of its ability to successfully suppress agriculture pests. The effectiveness of several biological control programmes was negatively affected by adverse environmental conditions. In particular, the variations in temperature during the course of a day strongly affect the performance of parasitoids reared in a laboratory for long periods (Firake & Khan, 2014). Because laboratory strains of parasitoids are usually reared under constant conditions, their effectiveness after release might be affected by the fluctuations in temperature they experience. Determination of the optimal thermal requirement for the development of a parasitoid is a fundamental aspect of biological control programmes against pests and is important for its laboratory production and field release. A previous study indicates that the optimum temperatures for the reproduction and development of adult D. longicaudata reared in larvae of B. dorsalis range from 24 to 27°C (Liu et al., 2012), but previously the effects of temperature on the immature stages of parasitoids were neglected. The activities of three antioxidant enzymes in oriental fruit fly larvae previously parasitized by D. longicaudata were considered to be a measure of the ability of parasitized hosts to simultaneously combat two different challenges, specifically parasitization and variations in temperature. Therefore, we studied the physiological responses of larvae of B. dorsalis and their parasitoids to keeping them at different temperatures for different periods of time.

A previous study indicates that the activity of SOD induced by varying the temperature protect fruit flies from...
heat stress (Jia et al., 2011). In the present study, the activity of SOD was significantly higher at 31°C 24 h after parasitization than after the same period of time in any of the other temperature treatments. Moreover, the activity of SOD was significantly lower in the other temperature treatments compared with that recorded at 25°C, probably because these unusual rearing temperatures suppressed the defensive antioxidant system of the cells, for example, by limiting the reduction of the superoxide radicals generated by ROS (Heck et al., 2003; Polte & Tyrrell, 2004). SOD removes superoxide anions generated by increased mitochondrial activity by dismutation into oxygen and H₂O₂, and then the H₂O₂ is converted stepwise into water and oxygen (Abele et al., 1998; An & Choi, 2010). Nevertheless, in our study, the levels of CAT were higher than that of SOD, which indicates that H₂O₂ was produced by other processes during thermal stress.

CAT is a light-sensitive antioxidant enzyme that is a crucial constituent of the insect antioxidant system; it is the sole enzyme responsible for converting H₂O₂ into water and oxygen because insects lack selenium-dependent glutathione peroxidase, which is a scavenger enzyme present in other organisms (Ahmad & Pardini, 1990). CAT is directly regulated by the amount of H₂O₂ in a cell (Lesser, 2006). CAT efficiently eliminates H₂O₂ only at high cellular concentrations (Ahmad et al., 1991; Ahmad, 1992). CAT activity was lower after 4 h than after other intervals of time, therefore, it is likely that CAT scavenges H₂O₂ during the later stages of parasitization by D. longicaudata. Our results indicate that the activity of CAT in parasitized larvae of B. dorsalis increased after 48 h of exposure to high temperatures (28, 31°C), similar to the increased activity of SOD, as shown in Fig. 3. In addition, the overproduction of CAT at high temperatures resulted in an enhanced elimination of H₂O₂, thereby preventing oxidative damage. Earlier studies indicate that the expression of the CAT gene in Drosophila melanogaster is strongly correlated with longevity (Orr & Sohal, 1994) and a reduction in the activity CAT or interruption in the expression of the CAT gene can result in death after emergence (Griswold et al., 1993; Orr & Sohal, 1994).

POX is also responsible for breaking down of H₂O₂ in organisms and their high tolerance of stress (Lee et al., 2005). The highest activities of POX were recorded after 5 and 24 h in all temperature treatments except 31°C, which may indicate that POX was mainly active after 5 and 24 h. At 31°C, a significant increase in the activity of POX was recorded after 24 h of exposure, whereas at 19°C, there was a remarkable decline in the activity of POX 48 h after parasitization. A previous study reports that a negative feedback from excess substrate due to oxidative changes can decrease enzyme activity (Tabatabaie & Floyd, 1994). The response of POX to changes in temperature deserve further evaluation.

A previous study reports that the eggs and larvae of parasitoids are more susceptible to temperature shocks than pupae (Firake & Khan, 2014). Hence, we reared the immature stages of D. longicaudata under different temperature conditions in order to improve the effectiveness of this parasitoid by acclimatizing it to the variable environment it will experience in the field. We considered that the activ-
ity of three major antioxidant enzymes (CAT, POX, SOD) indicates the ability of parasitized hosts to combat stresses and determined a preliminary temperature strategy (see Table 2) for rearing D. longicaudata-infected B. dorsalis larvae. Due to parasitization by D. longicaudata, the developmental period of the larvae of B. dorsalis was prolonged to 5.5 days, which may be attributed to the different metabolic conditions (Wang & Messing, 2003). Exposure to temperature treatment 1 (4 h at 28°C, 5 and 48 h at 25°C, 49 and 72 h at 31°C) had a significantly positive effect on percentage of eggs that hatched, pupal weight, percentage emergence and parasitoid longevity compared with the constant temperature treatment. The lower percentage emergence recorded in the control and temperature treatment 2 (48 h at 25°C, 49 and 72 h at 31°C), 3 (4 h at 25°C, 5 and 24 at 28°C, 25 and 72 h at 31°C) and 4 (24 h at 25°C, 25 and 72 h at 31°C) might be due to a higher mortality of the immature stages of the parasitoid (Chihrane & Laugé, 1996). The shrinkage or desiccation of host larvae may result in the development of weak and female parasitoids. Regardless of the undesirable sex ratio (female/male ratio below 1.00), temperature treatment 1 was accepted as the optimum temperature treatment for rearing immature parasitoids. However, the strong reduction in the number of females recorded in temperature treatment 1 is an important caveat.

A previous study indicates that species that perform well in the laboratory are more likely to do so in the field (Pak & Lenteren, 1986). However, this study was done under laboratory conditions and the effects of changing the temperature during the immature development of the parasitoid on the performance of parasitoids in the field needs to be evaluated. Moreover, the connection between the activity of antioxidants in fruit fly larvae subject to stress, such as parasitization and changes in temperature, and the development of the parasitoid inside a host larva is poorly understood and needs further investigation. Our experiments aimed to provide guidance for improving the mass rearing of braconid parasitoids by varying the temperature at which they are reared.

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