# Thermal requirements of *Aprostocetus hagenowii* (Ratzeburg, 1852) (Hymenoptera, Eulophidae) reared in oothecae of *Periplaneta americana* (Linnaeus, 1758) (Blattaria, Blattidae)

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## Resumo

Exigências térmicas de *Aprostocetus hagenowii* (Ratzeburg, 1852) (Hymenoptera, Eulophidae) criados em ootecas de *Periplaneta americana* Linnaeus, 1758 (Blattaria, Blattidae). O objetivo deste estudo foi determinar as exigências térmicas de *Aprostocetus hagenowii* em ootecas de *Periplaneta americana*, bem como a influência das diferentes temperaturas na biologia do parasitoide. Ootecas com idade máxima de oito dias e peso variando entre 0,09 e 0,10 g foram individualizadas em placas de Petri com um casal de *A. hagenowii*, ficando expostas ao parasitismo. Parasitoides e ootecas permaneceram juntos por 24 h a 25°C; após esse período, os casais foram retirados e as placas contendo ootecas foram transferidas para câmaras às temperaturas de 15, 20, 25, 27, 30 e 35°C. Para cada temperatura foram realizadas trinta réplicas. A duração do ciclo (ovo-adulto) de *A. hagenowii* apresentou uma tendência inversa à temperatura. A maior viabilidade de emergência do parasitoides. A Tb foi de 11,47°C e K foi de 613,5 graus-dia. Os resultados obtidos fornecem informações básicas para planejar a criação de *A. hagenowii* em laboratório, além de apresentar indícios sobre a dinâmica populacional desse parasitoide no ambiente.

Palavras-chave: Controle biológico; Parasitoide de ootecas; Temperatura

## Abstract

This study aimed to determine the thermal requirements of *Aprostocetus hagenowii* in oothecae of *Periplaneta americana*, as well as the influence of different temperatures on the biology of this parasitoid. Oothecae with maximum age of eight days and weight ranging from 0.09 to 0.10 g were individualized in Petri

plates with a couple of *A. hagenowii*, being exposed to parasitism. Parasitoids and oothecae stayed together for 24 h at 25°C; after this period, the couples were removed and the Petri plates containing oothecae were transferred to chambers at the temperatures of 15, 20, 25, 27, 30, and 35°C. Thirty replicates were conducted for each temperature. The cycle length (egg-adult) of *A. hagenowii* presented an inverse tendency to the temperature. The highest viability of emergence of this parasitoid (70%) found in this study was at the temperature of 25°C and the lowest viability (50%) occurred at 30°C. At the temperature extremes tested (15 and 35°C) no emergence of parasitoids was observed. Bt was 11.47°C and K was 613.5 degrees-day. The results obtained provide basic information to plan the rearing of *A. hagenowii* in laboratory, and they also present traces on the population dynamics of this parasitoid in the environment.

Key words: Biological control; Oothecae parasitoid; Temperature

The American cockroach, *Periplaneta americana* (Linnaeus, 1758) (Blattaria, Blattidae), is a highly synanthropic species, omnivore, which is elusive during the day and invades households, hospitals, and industrial facilities at night. It's an important mechanical vector of several pathogens, such as viruses, bacteria, fungi, cysts of protozoa, and helminths (eggs and larvae) (CLOAREC et al., 1992; THYSSEN et al., 2004). Besides, *P. americana* is associated to allergic reactions and it can cause health problems ranging from dermatitis to anaphylactic shock (LOPES et al., 2006), something which indicates its importance to public health.

The biological control of insects may constitute an alternative by reducing the damage caused by pests without affecting non-target insect populations. Parasitoids are among the options for biological control of *P. americana*. Lebeck (1991) reviewed hymenopteran natural enemies of cockroaches, focused on biological control, and this author pointed out eight species (predators and parasitoids), especially *Aprostocetus hagenowii* (Ratzeburg, 1852) (Hymenoptera, Eulophidae), a parasitoid with potential for reducing the population of this cockroach species.

*Aprostocetus hagenowii* is very adaptable to various climate conditions, it's found worldwide, in different geographic areas, such as Australia, India, Saudi Arabia, Taiwan, Trinidad and Tobago, United States, and Japan (NARASIMHAN; SANKARAN, 1979). In Brazil, it has been observed parasitizing oothecae of *P. americana* in Rio de Janeiro (DE SANTIS, 1980) and Rio Grande do Sul (CÁRCAMO et al., 2009).

Knowledge on the thermal requirements of parasitoids may be used to manage their population in the laboratory (PARRA, 1997), in order to determine the optimal temperature for their development and to synchronize the rearing of hosts and parasitoids (PRATISSOLI; PARRA, 2000).

This study aimed at identifying the thermal requirements of *A. hagenowii* in oothecae of *P. americana* and the effect of different temperatures on the biology of this parasitoid.

The adult specimens of *P. americana* and *A. hagenowii* were collected at the facilities of the Universidade Federal de Pelotas ( $31^{\circ}48^{\circ}S$ ,  $52^{\circ}25^{\circ}O$ ) and maintained in incubators at  $25^{\circ}C \pm 2^{\circ}C$ ,  $70\% \pm 10\%$  relative humidity (RH) and 12 h photoperiod. Cockroaches were fed with meat meal and sugar (1:1) and water *ad libitum*. The *A. hagenowii* specimens were fed with a solution of water and 20% of sugar.

For rearing of the parasitoids, oothecae of *P. americana* were exposed to 24 h in the glass tubes with *A. hagenowii* females. After this period, oothecae were individually placed at test tubes until the emergence of insects for the maintenance of the colony.

To standardize the experiments, the maximum age of oothecae was eight days. This time was chosen having a pilot experiment as a basis, which indicated that the characteristics of the progeny of *A. hagenowii* weren't affected using oothecae of up to 10 days of age at a temperature of  $25^{\circ}$ C.

The weight of oothecae ranged between 0.09 and 0.10 g. Oothecae were individually placed at covered Petri plates with 5 cm diameter, with a cotton ball embedded in a solution of water and 20% of sugar. Newly emerged *A. hagenowii* couples collected during mating were transferred to Petri plates (one couple per dish) containing one ootheca of *P. americana* and

maintained for 24 h in an incubator. After this period, couples were removed and Petri plates with oothecae were transferred to incubators at 15, 20, 25, 27, 30, and  $35^{\circ}$ C,  $70\% \pm 10\%$  RH and 12 h photoperiod. Each experiment temperature had thirty replicates, which were examined daily for the emergence of parasitoids or eclosion of nymphs. After 120 days, if neither of these events occurred, oothecae were dissected and examined under stereomicroscope for determining the presence or absence of parasitism.

The experimental design was randomized with 6 treatments and 30 replicates. The effect of the temperature on the emergence of parasitoids was assessed using generalized linear models (GLM) with quasipoisson distribution to correct the overdispersion of data. The influence of temperature in the development rate was evaluated with a linear regression. The significance of all tests was set at p < 0.05. Analyses were carried out with the statistical software R (R DEVELOPMENT CORE TEAM, 2010) following the suggestion of Crawley (2007).

The determination of the thermal requirements of *A. hagenowii*, the base temperature (Bt) and the thermal constant (K) were calculated through the hyperbole method (HADDAD et al., 1999), based on the cycle length (egg-adult) at the tested temperatures.

The linear regression analysis of the average developmental rates and the temperatures tested resulted in the model 1/D = -0.0187+0.00163x, (F = 1.69 = 270.5; p < 0.001; r<sup>2</sup>= 0.794), indicating that this variable strongly influences on the development of *A. hagenowii* and that the development speed is inversely associated to temperature.

The Bt from egg to adult life of this parasitoid in oothecae of *P. americana* was 11.47°C and the thermal constant was 613.5 degrees-day. The results weren't significantly different from those estimated through the chi-square test ( $x^2 = 0.1405$ ; (0.001)). The Bt found in this study is similar to that found by Bressan-Nascimento et al. (2010) for males (Bt = 12.7°C) and females (Bt = 12.8°C) of *Evania appendigaster* (Linnaeus, 1758) (Hymenoptera, Evaniidae), also an ootheca parasitoid. Despite a Bt of 11.47, the emergence of parasitoids wasn't observed at 15 and 35°C, only the eclosion of nymphs, unlike the results found by Narasimhan (1984). This author reported that *A. hagenowii* can develop at 15°C, taking around 61 days to reach the adult stage. These variations in viability, regarding the upper and lower temperature limits, might be due to specific features of populations from different geographic regions. The highest viability (70%) was found at the temperature to rear this parasitoid, since it's the closest to the temperature regarded as optimal for artificial diets (75%), according to Parra (2002).

The life cycle length (egg-adult) obtained in this study (Table 1) is similar to that found by Hagenbuch et al. (1988), 35.77 days at 27°C, and by Narasimhan (1984), who reported a total cycle of 44±4 and 33±5 days, for the temperatures of 25°C and 30°C, respectively. Narasimhan (1984) also reported another ootheca parasitoid, *Aprostocetus asthenogmus* (Waterston, 1915) (Hymenoptera, Eulophidae), which has a developmental period similar to that of *A. hagenowii*, with a life cycle length (egg-adult) between 36 and 52 days at 26°C.

The development time of *E. appendigaster*, also a parasitoid of oothecae of *P. americana*, is similar to that observed for *A. hagenowii* (BRESSAN-NASCIMENTO et al., 2010). However, *A. hagenowii* is a gregarious parasitoid and, thus, it requires fewer hosts to produce the same number of *E. appendigaster* individuals, making mass rearing more feasible. Another advantage pointed out by Kumarasinghe and Edirisinghe (1987) is that when larvae of these two species compete, only *A. hagenowii* emerges, except in oothecae parasitized by *E. appendigaster* for more than 32 days.

According to Pratissoli and Parra (2000), the life cycle length (egg-adult) of some parasitoids of the genus *Trichogramma* Westwood, 1833 (Hymenoptera, Trichogrammatidae) don't depend only on temperature, but also on the species or lineage adaptation and on the host. This probably doesn't occur in *A. hagenowii*, as the periods observed in this study with oothecae of *P. americana* are similar to those observed by Hagenbuch et al. (1988), using oothecae of other cockroaches species as hosts. TABLE 1: Mean  $\pm$  standard error life cycle length (egg-adult), variation in the developmental period (in days), sex ratio, mean number of parasitoids per ootheca, and percentage of emergence of *Aprostocetus hagenowii* reared in *Periplaneta americana* oothecae, at different temperatures;  $70 \pm 10\%$  RH; 12h photophase.

<b>Temperature</b> (°C)	Life cycle length (days)	Variation in the developmental period (days)	Sex ratio	Number of parasitoids per ootheca	Percentage of emergence (%)
15	*	*	*	*	*
20	$70.31 \pm 15.02$	51 - 86	$0.870 \pm 0.0457$	$69.11 \pm 21.13$	60.00
25	$46.43 \pm 5.43$	42 - 50	$0.833 \pm 0.0573$	$63.86 \pm 25.32$	70.00
27	$38.12\pm10.03$	25 - 49	$0.834 \pm 0.0351$	$66.00 \pm 19.03$	56.67
30	$32.13 \pm 4.81$	28 - 37	$0.870 \pm 0.0436$	$62.13 \pm 20.73$	50.00
35	*	*	*	*	*

\* No parasitoid emergence.

The developmental period length of *A. hagenowii* is also similar to the embryogenesis period found by Bressan-Nascimento et al. (2008) for the host used in this study. This can make difficult mass rearing and release of this parasitoid, since it isn't possible to separate development from embryogenesis based on the development time of parasitoid or host using temperature as a tool.

The sex ratio didn't significantly vary with the independent variable (Table 1), suggesting that other factors are involved. For this parasitoid species, two other factors influence on the number of parasitoids per ootheca and the sex ratio: the ootheca size and the number of parasitoids developing in each host (NARASIMHAN, 1984). The sex ratio remained high at all temperatures. This result is very important, because the female parasitoids are responsible for the progeny production, thus, they're responsible for suppression of the target species populations.

The total number of parasitoids per ootheca wasn't significantly different among the temperatures examined. The average number of parasitoids per ootheca ranged from 62.13 to 69.11, at the temperatures of 30°C and 20°C, respectively ( $x^2 = 5202$ ; p = 0.2; df = 119). This result is in accordance with that observed for the sex ratio, are it's important because allows focusing on the production and release of parasitoids taking into account just the viability of parasitism and the developmental period.

The best temperature for rearing *A. hagenowii* is 25°C with emergence of 70% of parasitoids with no

change in the number of parasitoids or sex ratio. The shortest period occurred at the temperature of 30°C, but this was the temperature showing the lowest parasitism viability (50%). At the extreme temperatures tested (15 and 35°C), no parasitoid emergence was observed. Tb was 11.47°C and K was 613.5 degree-days. Knowing these biology aspects of this parasitoid is very important to plan the rearing of these insects in laboratory and also to present evidence on their population dynamics in the environment.

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