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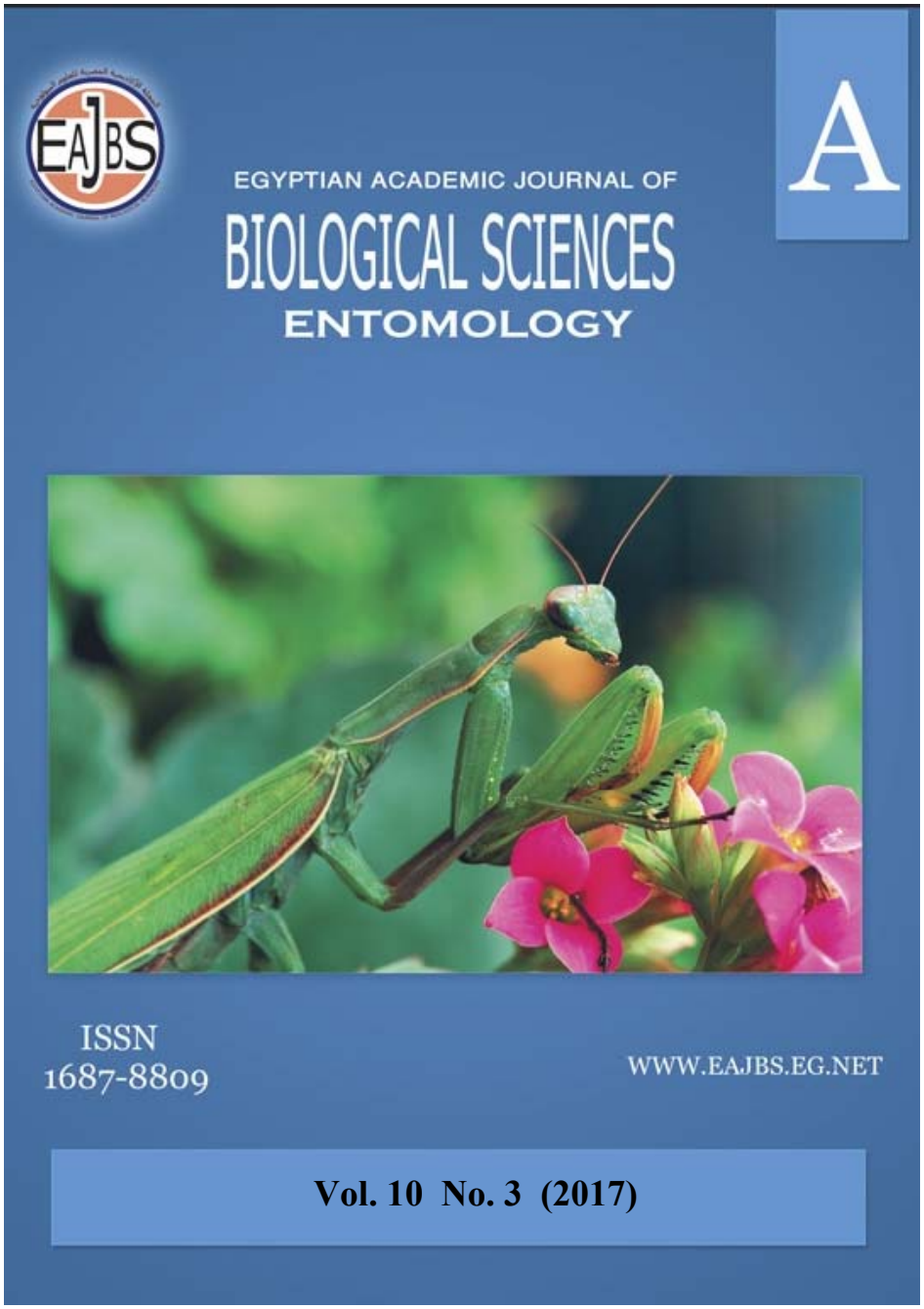

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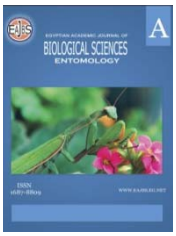

Threshold Temperatures and Thermal Requirements for the Development of the Olive Leaf Moth; *Palpita unionalis* Hbn. (Lepidoptera: Pyralidae)

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ABSTRACT

Olive leaf moth, *Palpita unionalis* (Hubner) (Lepidoptera: Pyralidae) is one of the dangerous pests attacking sapling olive trees and nurseries, tender shoots and ripening fruits inducing economic losses. Accordingly, great efforts were dedicated to reduce its infestation. The current study aimed to calculate temperature thresholds (t_0) and accumulated heat units (dd's) for each stage of this pest as a primary step for developing a forecasting system that will help to define the most precise time for different control programs. The insect was reared under three constant temperatures (17, 22 and 27°C each ± 1°C). The time required for development through egg, larva, pupa and pre-ovipostion increased at lower temperatures. The lower thresholds of development (t_0) were 8.39, 11.46, 13.38 and 12.70°C for eggs, larvae, pupae and pre-oviposition period, respectively. The average accumulated heat units required for their development were 61.07, 238.81, 113.68 and 38.17 degree-days. On the other hand, the lower threshold of development (t_0) to complete a generation was 12.04°C and the average accumulated heat units required for its development was 443.07 degree-days.

INTRODUCTION

Olive leaf moth, *Palpita unionalis* (Hübner) (Lepidoptera: Pyralidae) is one of the commonly distributed lepidopterous olive pests throughout the whole olive cultivating countries especially those at the Mediterranean basin (Noori and Shirazi, 2012). Various field and laboratory issues regarding *P. unionalis* had been considered in different countries including its worldwide distribution (Ghoneim, 2015), host range (Mazomenos *et al*., 2002), morphological characteristics (Santorini and Vessiliana, 1976 and Stefanos, 2003), life cycle of its stages (Yilmaz and Genc, 2012), life table parameters (Mansour *et al*., 2016) and population fluctuation (Kovanci *et al*., 2006). Induced losses due to *P. unionalis* attack was the main theme considered by a lot of reviewers and authors due to its direct influence on the economic return of the farmers. In this regard Yilmaz and Genc (2013), Athanassiou *et al*., (2004) and Kumral *et al*., (2007) pointed out the significant damages of *P. unionalis* larval stage on sapling olive trees and nurseries, tender shoots and ripening fruits.

Arambourg (1986) and Lopez-Villalta (1999) stated that there were 90% of the total olive leaf area and 30% of olive fruit yield got injured due to *P. unionalis* larval attack. Due to its polyphagous behavior, considerable reduction in the jasmine oil production was stated in France as a negative feedback from the invasion of its larval stage on leaves and flower buds of jasmine plants (Gargani, 1999). Accordingly, great efforts were dedicated to face this economic pest and to reduce its infestation.

For environmental cleanliness and public health considerations, integrated pest management (IPM) strategies, nowadays, acquire much attention in pest control programs at which augmentation of bio-candidates plays a vital role in such IPM programs (Jarrahi and Safavi, 2016). Out of environmental factors, temperature is the vital attribute that responsible for deriving field scenarios of cold-blooded arthropods (their biological activities and population dynamic regulation). So, for adjusting the emergence pattern of the target pest stages (either egg or larval stages) to meet the proposed augmentation schedule of bio-candidates, temperature dependent models should be designed for monitoring the developmental growth of the intended pest (Guillen *et al*., 2007). Where, according to Wagner *et al*., (1984) and Dahi *et al*., (2009) each insect has its own temperature limits or thresholds. The lower threshold under which no development occurs and the upper one (upper cut off) above which no development also occurs. In this regard the linear relation between the rate of development (1/developmental time) and different temperature regimes became highly sounded in detecting both the lower developmental threshold and the number of accumulated heat units that called degree-days (dd's) for vast range of insect and acarine species with the aim of optimizing the control programs targeted such pests (Alois, 1996 and Wagner *et al*., 1984). To our knowledge, since there is no any documented data regarding the thermal requirements of the olive leaf moth, so the current study has been proposed to find out the temperature threshold (t_0) and the accumulated heat units (dd's) for each stage of this pest as a primary step for predicting field phenology model of the olive leaf moth depending on the calculated thermal units.

MATERIALS AND METHODS

The laboratory colony of *P. unionalis* was initiated with specimens collected from olive orchards in Matrouh Governorate. Infested twigs were left in 2-pound plastic jars, daily provided with fresh olive leaves and kept in growth chamber at a constant temperature of 27 ±1°C. Adults were collected shortly after exclusion, transferred to 5-pound plastic jar and allowed to be sexed. Moths were provided with 10 % sugar solution for feeding and olive twigs for oviposition. Moths were transferred to new jars as deposited eggs started to hatch. Neonate larvae were frequently provided with tender olive leaves until pupation.

For the study, more than 800 *P. unionalis* eggs were incubated at three constant temperatures (17, 22 and 27°C) in order to determine the rate of development. Daily observation was continued to record the time of egg hatch and number of hatched eggs. Incubation period as well as rate of egg development ([1/developmental time (t)]*100) and hatching rate was estimated. Daily observation of neonate larvae was undergone to record larval mortality and larval developmental period. Larval developmental rate and larval survival rate were calculated. Newly formed pupae were collected and observed for adult emergence. After being sexed, the newly emerged female moths of each group were observed to record pre-oviposition period after provided with 10 % sugar solution for feeding. The developmental rate was

calculated for pupae and moths through their pre-oviposition period. Pupal survival rate and sex ratio of emerged moths were calculated. Duration of the whole generation (egg-end of pre-oviposition period) was also calculated. Differences in each measured parameter under different constant temperatures was examined by one-way analysis of variance (ANOVA) followed by Duncan's multiple range test using SPSS software (IBM SPSS Statistics for Windows, 2013).

In order to calculate the theoretical developmental thresholds (t_0) and the accumulated thermal units (K), the regression formula was used:

$$y = a + b\,x$$

$$(t_0) = -a / b;\ K = 1 / b$$

Where: y= developmental rate of a given stage; x= temperature in degrees centigrade; (a): constant term; (b): regression coefficient; (t_0): lower threshold of development and (K): thermal units.

On the other hand, the thermal units those required to complete each stage development was determined according to equation of the thermal summation **(Blunk, 1923):**

$$K = y\,(T - t_0)$$

Where: K = thermal units (degree-days (dd's)); y = developmental duration of a given stage; T= temperature in degree centigrade and t_0 = lower threshold of development.

RESULTS

Data of the current study exhibited fluctuated patterns of the developmental schedule of *P. unionalis* stages (eggs, larvae, pupae) as well as pre-oviposition period and the duration of the generation that experienced different temperatures. Table (1) revealed that all measured durations at low temperature (17°C) exhibited significant variation upon compared with that measured at higher temperatures. Whereas the higher subjected temperatures (22, 27 °C) showed non-significant differences when measuring the same duration.

Table 1: Development of *P. unionalis* (days ± SE) under different constant temperatures.

Temp. (°C)	Incubation period	Larval duration	Pupal duration	Pre-oviposition period	Duration of generation
17	7.82±0.13^{a}	43.00±3.33^{a}	27.03±0.42^{a}	9.67±3.53^{a}	87.18±0.13^{a}
22	3.88±0.13^{b}	22.71±1.11^{b}	15.51±0.17^{b}	3.69±0.61^{b}	45.79±0.13^{b}
27	3.39±0.17^{c}	15.36±0.69^{c}	8.04±0.13^{b}	2.70±0.19^{b}	29.49±0.17^{c}

The values having the same letters vertically are non significant for difference.

Through considering the incubation periods, it was noteworthy that eggs subjected to low temperature (17°C) took about double time (7.82 ± 0.13) to hatch comparing with those at higher temperatures which differed significantly (F= 373.5; P= 0.00). As a holistic conclusion, the other recorded durations behaved a similar trend. Where, values of larval duration (F= 44.918; P=0.000), pupal duration (F= 1375.87; P=0.000), pre-oviposition period (F= 10.752; P=0.000) and duration of generation (F= 5000.913; P=0.000) detected at 17°C were significantly differed from its fellows measured at the higher applied temperatures.

In the same context, both survival rate and sex ratio of the emerged adults also exhibited different responses upon subjected to the proposed temperatures (Table 2). The rate of egg hatching at 17, 22 and 27°C were categorized at the same statistical

group (F= 5.62; P=0.003). Pupation and adult emergence rates were grouped under two statistical groups. Both rates at 17°C were significantly differed from that recorded at the remainder temperatures (F=8.044; P=0.001 for pupation rate and F=3.84; P=0.02 for emergence rate). The last parameter, sex ratio, showed non-significant differences at the examined temperatures (F=0.974; P=0.419).

Table 2: Survival rate of *P. unionalis* and sex ratio of emerged adults under different constant temperatures.

Temp. (°C)	Hatch rate	Pupation rate	Emergence rate	Sex ratio
17	0.83±0.05[a]	0.89±0.07[a]	0.66±0.09[a]	0.53±0.12[a]
22	0.93±0.04[a]	0.40±0.10[b]	0.85±0.03[b]	0.61±0.67[a]
27	0.75±0.08[ab]	0.47±0.07[b]	0.83±0.05[b]	0.51±0.33[a]

The values having the same letters vertically are non significant for difference

Both lower threshold of development (t_0) and accumulated heat units (dd's) were calculated at the subjected thermal regimes (Table 3). In this regard, about 67.27, 52.79 and 63.16 accumulated heat units (dd's) were required for the incubation period at 17, 22, 27°C, respectively.

Table 3: Thermal requirements of *P. unionalis* under different constant temperatures.

Temp. **(°C)**	Temperature threshold (t_0) and degree-day units (dd's) for									
	Incubation period		Larval duration		Pupal duration		Pre-oviposition period		Generation	
	t_0	dd's	t_0	dd's	t_0	dd's	t_0	dd's	t_0	dd's
17	8.39	67.27	11.46	238.31	13.38	97.82	12.70	41.56	12.04	432.16
22		52.79		239.46		133.67		34.34		455.92
27		63.16		238.65		109.55		38.61		441.13
Average		**61.07**		**238.81**		**113.68**		**38.17**		**443.07**

The lower threshold of egg development (t_0) value was 8.39°C according to the regression equation y = 1.67x – 13.99 (Fig. 1A). Thermal units required for both larvae and pupae were 238.31, 239.46, 238.65 and 97.82, 133.67, 109.55 at 17, 22, 27°C, respectively. The lower threshold of development (t_0) for the same stages were 11.46 and 13.38°C according to the regression equation y = 0.42x – 4.80 and y = 0.87x –11.69 (Fig. 1 B, C).

Regarding pre-oviposition period, 41.56, 34.34, 38.61 were calculated as thermal units required at the same temperatures. The lower threshold of development (t_0) was 12.70°C according to the regression equation (y = 2.67x – 33.90) (Fig. 1D). When calculating the thermal units (dd's) for *P. unionalis* generation, it was found to be 432.16, 455.92, and 441.13 at 17, 22 and 27°C, respectively.

The lower threshold of development (t_0) was 12.04°C according to the regression equation y = 0.22x – 2.71 (Fig. 2).

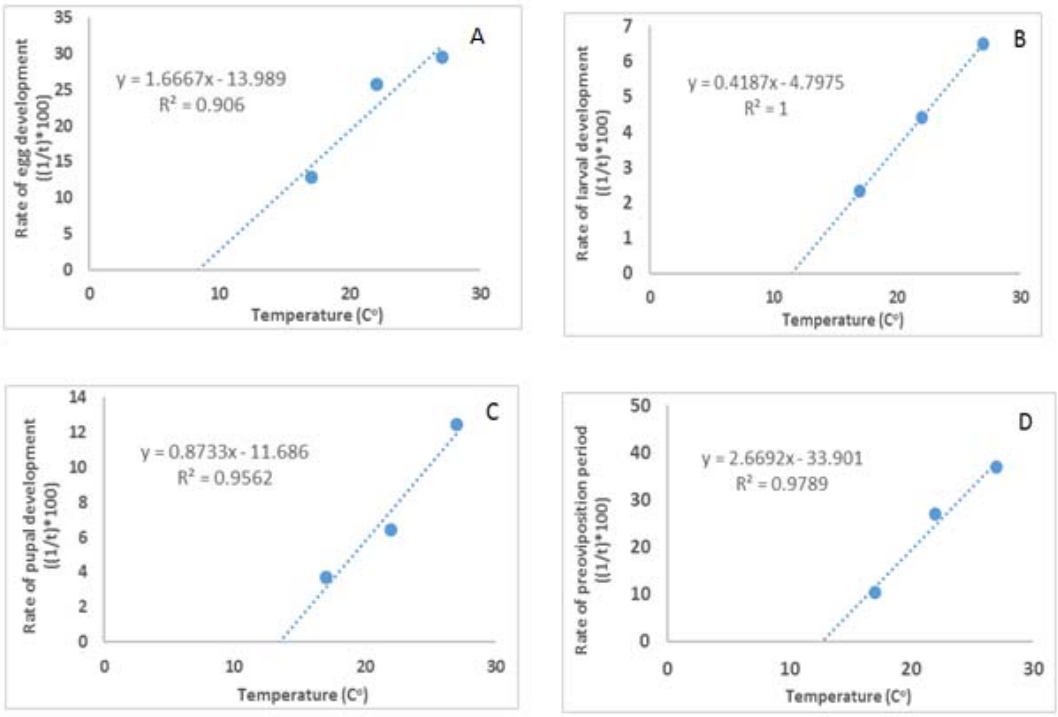

Fig. 1: Linear regression of *P. unionalis* developmental rate ((1/t)*100) during incubation (A), larval (B), pupal (C) and pre-oviposition periods (D) against temperature.

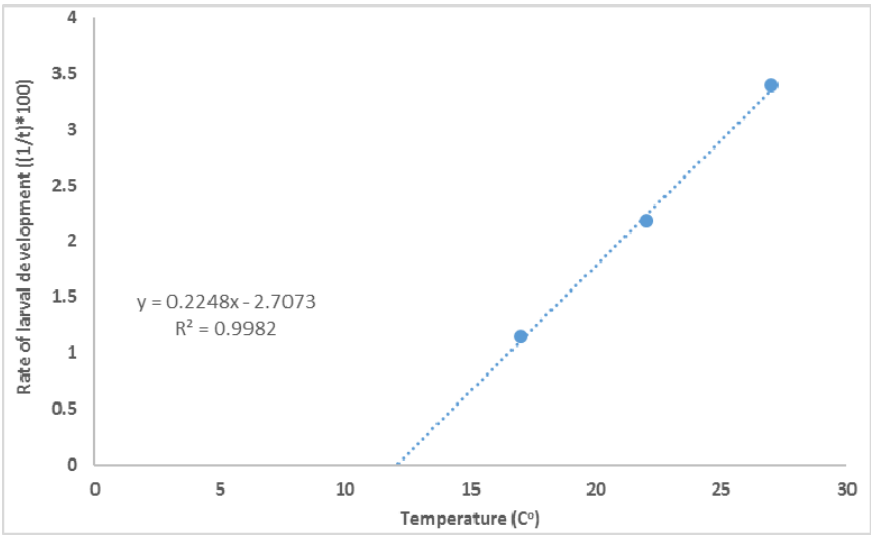

Fig. 2: Linear regression of *P. unionalis* developmental rate ((1/t)*100) against temperature.

DISCUSSION

Although there is no study considered the thermal requirements of the olive leaf moth; *P. unionalis* so far, many authors studied its biology at different temperatures. Fouda (1973) and Badawi *et al*. (1976) reported that egg stage varied from 3 days at 30°C to 12 days at 15°C. Moreover, the mean larval duration period ranged between 16 and 26 days at 30 and 20°C. The same author reported that at 35°C, all eggs failed to hatch and larvae died after the first moult. The pupal stage averaged 31, 17, 9 and 8 days for males and 23, 16, 8 and 7 days for females at 15, 20, 25 and 30°C, respectively. Pre-oviposition period was 3.5 days in average at

27°C. El Khawas (2000) estimated the incubation period to range between 1 and 3 days under 25°C, while the mean total larval duration lasted 15.8 days and the total developmental period averaged 23.4 days. Kumral *et al*., (2007) recorded 30 days developmental time of immature stages on olive leaves at 25°C. Khaghaninia and Pourabad (2009) estimated the mean developmental time from the egg to the adult at 27°C to be 34.9 days. Alavi (2010) found that at 25°C, eggs hatched after 2.5 days, the larval and pupal stages lasted for 21.5 and 8.6 days, respectively. Noori and Shirazi (2012) found the mean incubation, larval and pupal durations as 6, 22 and 8 days at 25°C. Yilmaz and Genc (2012) recorded 4, 23, 10 and 2 days for egg, larval, pupal and pre-oviposition durations at 24°C. The slight differences between authors and the present data (duration of generation was 30-87 days) could be attributed to the difference in the pest strains, the difference in olive varieties used for larval feeding or different temperature regimes used through laboratory studies.

Survival rate of different immature stages during the current work was represented by 0.75- 0.93, 0.40-0.89 and 0.66-0.85 for eggs, larvae and pupae, respectively. This was in synchrony with Kumral *et al*. (2007) who recorded survival percentage of immature stages as 73%. In the same regard, Rahhal (1972) recorded 78-95% hatchability percent and Fouda (1973) recorded 68, 94, 98 and 92 hatchability percent at 15, 20, 25 and 30°C. This was coincided with Loi (1990) who reported the hatched eggs to be 68 and 98% at 15 and 25°C. Later, the survival of larval stages was recorded as 60% while the survival rate of pupae was 82.8% at 24°C (Yilmaz and Genc, 2012). Sex ratio during the course of the present research was 0.51-0.61. These findings matched with other authors who recorded sex ratio to be 0.54 (Fodale and Mule, 1990), 0.53 (Fazel. and Azimizadeh, 2010), 0.46 (Yilmaz and Genc, 2012) and 0.40 (Noori and Shirazi, 2012).

The degree-day (dd's) is the most important thermal component that express the physiological time that required to complete a specific event in insect developmental schedule. The degree-days values of the considered parameters at Table (3) met reversible relationships with the applied thermal regimes. Such findings had been stated by several authors; Dahi *et al*., (2009) on *Agrotis ipsilon* and O'neal *et al*., (2011) on *Marmara gulosa,* Where, their obtained results clarified the importance of the relationship between growth and developmental patterns of the intended insect and its thermal requirements. Such ecological relationship could be exploited to optimize a precise population dynamic model that could act as a vital tool in any proposed control strategy. In the same concern, the output of the current study could be a valuable guideline for the management programs that dedicated against the olive leaf moth through precisely detecting the time of treatment to be matched with the target stage of the pest.

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ARABIC SUMMERY

الحدود و المتطلبات الحرارية لنمو دودة أوراق الزيتون الخضراء (حرشفية الأجنحة: الفراشات ذات الخرطوم).

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١- معهد بحوث وقاية النباتات – مركز البحوث الزراعية – الدقي – الجيزة - مصر.

٢- قسم وقاية النبات - مركز بحوث الصحراء – المطرية - القاهرة - مصر.

تعد دودة أوراق الزيتون الخضراء (حرشفية الأجنحة: الفراشات ذات الخرطوم) واحدة من الآفات الخطيرة التي تهاجم شتلات الزيتون، النموات الخضرية و الثمار مما يسبب الكثير من الخسائر الإقتصادية. وبالتالى فهناك الكثير من الجهود المبذولة للحد من انتشارها. ومن هنا تستهدف الدراسة الحالية حساب الحدود الحرارية، صفر النمو البيولوجى (t_0) والوحدات الحرارية المتجمعة لكل مرحلة من مراحل دورة حياة هذه الآفة كخطوة أساسية فى تطوير نظام للتنبؤ يساعد على تحديد الوقت الأكثر دقة لمختلف برامج المكافحة المستخدمة ضد الآفة. و فى أثناء ذلك تم تربية الآفة تحت ثلاث درجات حرارة ثابتة (17، 22 و 27 ± 1 درجة مئوية). تم حساب الوقت اللازم لنمو البيض، اليرقات، العذارى و فترة ما قبل وضع البيض عند درجات الحرارة المختلفة. لوحظت العلاقة العكسية بين الوقت اللازم للنمو و درجات الحرارة المستخدمة. تم حساب صفر النمو البيولوجى (8.39، 11,46، 13,38 و 12,70 °م) لكل من البيض، اليرقات، العذارى و فترة ما قبل وضع البيض على التوالى. وكان متوسط الوحدات الحرارية المتجمعة اللازمة لنمو كل طور 61.07، 238.81، 113.68 و 38.17 درجة يوم. من ناحية أخرى، كان صفر النمو البيولوجى للجيل الكامل من الآفة 12.04 درجة مئوية، وكان متوسط الوحدات الحرارية المتجمعة اللازمة للجيل الواحد 443.07 درجة-أيام.

الكلمات المفتاحية: دودة أوراق الزيتون الخضراء، بالبيتا يونيونااليس، الوحدات الحرارية المتجمعة (درجة-أيام)، صفر النمو البيولوجى (t_0).