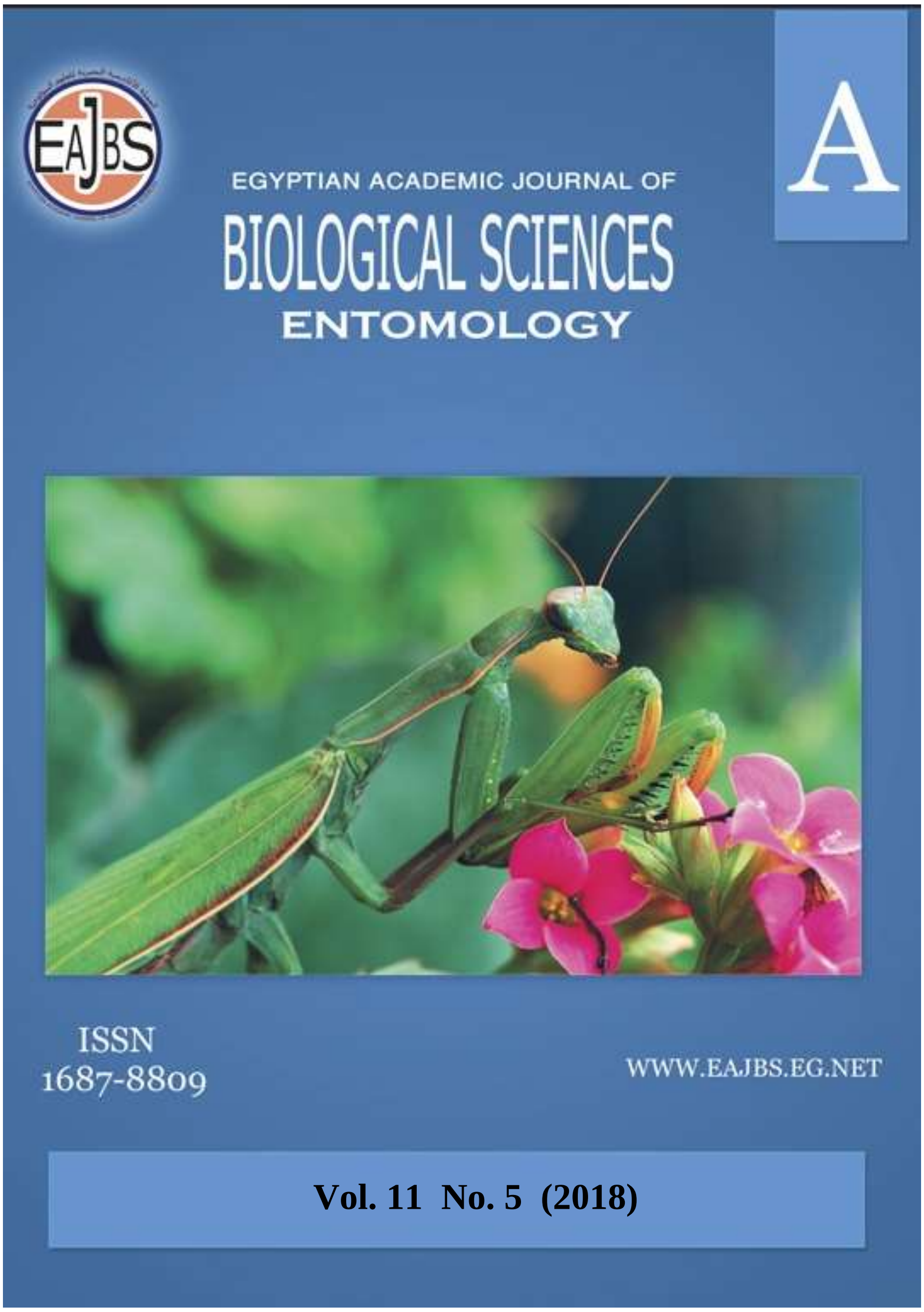

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The Relation Between Developmental Stages of the Predator, *Rodalia cardinals* (Mulsant) Reared on *Icerya purchasi* Maskell and the Required Thermal Units

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ABSTRACT

Laboratory experiments were carried out in the Insectary of Economic Entomology Department Faculty of Agriculture, Mansoura University study the relationship between the developmental stages of the predator, *Rodalia cardinals* (Mulsant) and the required thermal units at three constant temperatures, (15, 25 and 30˚C) on the third nymphal instar of *Icerya purchasi* Maskell. The results indicated that, the duration of the predator was longer at 15˚C. As temperature increased from 15 to 30˚C the developmental aspects of *R. cardinals* decreased. The lower thermal threshold for the different stages were 10.5, 8.5, 5.3, 5.2, 5.4, 6.9, 0.8 and 1.3˚C for egg stage, larval stage, pupal stage, preoviposition period, oviposition period, postoviposition period, adult female longevity and adult male longevity, respectively. The heat units required for the development of eggs was 56.2 DD unite, while for larvae, pupae, adult female and adult male were 211.3, 114.7, 969.3 and 664.4, consecutively.

The results revealed that, the longevity of the predator adult stage decreased with increasing the temperature degrees. The highest consumption rate per female was at 15 ˚C comparing with the other temperature degrees. The highest number of deposited eggs per female was 25 ˚C with the value of 443.71 eggs/ female. Obtained results provide essential information for predicting the field population of the predator *R. cardinals*, releasing time on certain mealybug species for controlling this pest and it's recommended to be an item of integrated pest management programs in Egyptian field designed to control certain mealybug species.

INTRODUCTION

Several mealybug species are pests of citrus, fruit trees, ornamental plants and grapevine in Egypt. The cottony cushion scale, *Icerya purchasi* Maskell, the Egyptian fluted mealybug, *Icerya aegyptiaca* (Douglas) and the seychellarum mealybug, *Icerya seychellarum* (Westwood) are important pests in many parts of the world especially in the tropical and subtropical regions. Their high harm is mainly due to the absence of effective entomophagous insects which could reduce their numbers (Abd Rabou, 2001; Esfandiari and Mossadegh, 2007; Abdel-Salam *et al.*, 2010 and Mohamed, 2013). The success of *Rodalia cardinals* (Mulsant) (Coleoptera: Coccinellidae) in controlling the cottony cushion scale, *I. purchasi* Maskell in the citrus groves of California in the 1880s and the subsequent liberation of this beetle in many other countries at the beginning of the 20th century (Bennett *et al.*,1985; Caltagirone and Doutt, 1989 and Mohamed *et al.*, 2013). Although observations of the life history of *R. cardinalis* suggest that this predator is specialized on Margarodidae, in reviewing the literature and specimen labels we found that there was only limited evidence of stenophagicity. Life cycle completion or feeding by *R. cardinalis* has been reported on several genera of Margarodidae (Causton *et al.*, 2004). In Egypt, few information were available on the influence of different temperatures degrees and prey types on the biological characteristics and life table parameters of the most important predators feeding on mealybug species. However, scant attention has been paid to the developmental time, consumption rate, longevity and fecundity of this predator to measure these parameters for mass rearing and release (Ragab, 1995 and Saleh *et al.*, 2017). Therefore, the objective of this investigation aimed to study the influence of different constant temperature degrees on the biological characteristics of this coccinellid predator *R. cardinalis* under laboratory conditions to control the cottony cushion scale because of its high reproduction rate, rapid development and host specificity (Quezada and Debach, 1973).

MATERIALS AND METHODS

Laboratory experiments were carried out in the Insectary of Economic Entomology Department, Faculty of Agriculture, Mansoura University, from the beginning of October 2016 tell the end September 2017 under three constant temperature degrees (15, 25 and 30 ˚C ±1). To obtain a culture from *Rodolia cardinalis* (Mulsant) a large number of this predator in the pupal stage were collected from ficus, *Ficus nitida* Thunb., guava trees, *Psidium guajava* L. and citrus trees which were found to be a heavily infested with *Icerya purchasi* Mask. and transferred to the laboratory until the emergence of the adults. Newly deposited eggs of this predator were divided into three groups; each group consisted of 50 eggs each group of the eggs was kept at one of the following constant temperature degrees 15, 25 and 30 ˚C ±1 as well as 70±5 % R.H.

A: Larval Experiments :

To avoid cannibalism , newly first larval instar of the predator from each group were individually in Petri dishes (10 cm diameters) and divided to three groups consisted of 20 larvae was used as a replicate and fed on *I. purchasi* nymphs. Apiece of filter paper was placed on the bottom of each Petri dish to provide a walking surface for the predator larvae. Known surplus numbers of the third nymph instar of I. *purchasi* species were offered and the devoured individuals were replaced daily for *R. cardinalis*. A small leaves from ficus or guava replaced daily as a food for the third nymphal instar of this mealybug species. Attached prey individuals were counted and recorded daily throughout the period of the larval instars.

B: Adult Experiments:

After emergence from the pupae, the predator adults were sexed and then introduced singly into a Petri dish. Know numbers of *I. purchasi* nymphs were offered daily on a ficus or guava leaflet to predatory adults. Counting and removing the undevoured nymphs in Petri dish were practiced before introducing the new nymph individuals. After five or six days of emergence, copulation took place and the two sexes were immediately separated and kept singly in the dishes. Daily numbers of laid eggs per predator female during its ovipositional period was counted.

In addition, the total number of eggs laid per predator female was estimated. The daily consumption throughout adult was calculated.

C: Accumulated Degree Days Calculation (ADD):

The lower developmental threshold (T_0) and the thermal constant (K) of *R. cardinalis* stages were estimated using the thermal summation model, which describes the relationship between the developmental rate of insects and the ambient temperature in a linear regression equation.

$$K\ (DD) = d\ (T - T_0)$$

Where K (DD) is the species (or stage-specific) thermal constant of the poikilothermic organism, T temperature, T_0 developmental zero temperature and d the duration of the stage by days. This thermal constant provides a measure of the physiological time required for the completion of a developmental process and is measured in degree days (DD).

One popular method of estimating the above parameters is to use a linearizing transformation of the above function by calculating the rate of development

$$DR = 1/d.$$

The linear degree day model or as the x-intercept method which is simply derived after growth rate fitting to a simple linear equation and then extrapolated to zero.

$$Y\ (DR) = a + bT$$

The lower theoretical temperature threshold is derived from the linear function as $T_0 = -a/b$ whereas 1/slope is again the average duration in degree days or thermal constant K. The Equation simply means that the thermal constant is a product of time and the degrees of temperature above the threshold temperature. (Campbell *et al.*, 1974; Haddad *et al.*, 1999; Bergant andTrdan, 2006 and Damos & Savopoulou-Soultani, 2012).

D: Statistical Analysis:

Data were statistically analyzed by using COSTAS Computer Program (2005).

RESULTS AND DISCUSSION

Egg Stage:

Data presented in Table (1) indicated that the incubation period of *R. cardinalis* was 9.68 days at 15 ˚C, with increasing temperature the developmental time of the egg stage decreased to 5.21 days at 25˚C and 2.53 days at 30˚C.

Data presented in Table (2) showed that the rate of development of the different life history stages in relation to temperature expressed by the linear regression equation. According to the regression line equation the lower developmental threshold of *R. cardinalis* eggs was10.5 ˚C and accumulated degree days (ADD) required for egg complete development was 56.2 DD unite.

Grafton-Cardwell *et al.* (2005) recorded the effects of temperature from 25 to 37 ˚C on the egg hatching of *R. cardinalis*, they mentioned that eggs held at 37 ˚C failed to hatch. There was no egg eclosion at 10 ˚C. No mortality occurred for the 22 and 26 ˚C temperatures. Survival was reduced to 79% at 18 ˚C and 63% at 14 ˚C. Eggs showed decreasing periods of days to eclosion from 15.7 days at 14 ˚C to 4.6 days at 26 ˚C.

Larval Stage:

Data illustrated in Table (1) mentioned that the larval stage development took 30.08 days at 15 ˚C and decreased to 14.32 days at 25˚C and 9.40 days at 30˚C.

Data tabulated in Table (2) showed that the lower developmental threshold of *R. cardinalis* larva was 8.50 ˚C and accumulated degree days required for the larva to complete development was 211.3 DD unite.

Grafton-Cardwell *et al.* (2005) indicated that larvae required 11.8, 6.7, 6.8, and 4.6 days for 50% completion of each of the four larval instars. In contrast, at 26 ºC larvae required 3.6, 1.5, 1.1, and 1.2 days to 50% completion of each of the four larval instars.

Table (1): Biological parameters durations (mean ± SE) in days of *Rodolia cardinalis* (Mulsant) reared on *Icerya purchasi* Mask. at constant temperatures (15, 25 and 30˚C) ± 1˚C, 65 ± 5 RH%

Temperatures / Biological parameters	15 ˚C	25 ˚C	30 ˚C	P	LSD
	Mean ± SE	Mean ± SE	Mean ± SE		
Egg incubation period	9.68	5.21	2.53	0.0001**	1.05
Larval stage	30.08	14.32	9.40	0.0002**	7.24
Pupal stage	10.42	7.08	4.19	0.0001**	9.17
Total developmental time	50.18	26.61	16.12	0.0001**	1.05
Preoviposition period	10.60	5.53	4.20	ns	
Generation	60.78	32.14	20.33	0.0001**	1.06
Oviposition period	38.75	29.68	22.19	0.0001**	2.31
Postoviposition period	16.86	6.23	5.76	0.0002**	1.05
Adult female longevity	66.21	41.44	32.15	0.0001**	1.52
Adult male longevity	45.96	28.57	23.92	0.0002**	1.40

Table (2): Lower threshold temperature (T_0), duration (D) and accumulated degree days (ADD) of different stages of *Rodolia cardinalis* (Mulsant) reared on *Icerya purchasi* Mask.

Temperatures / Stages	15 ˚C			25 ˚C			30 ˚C			Average of ADD
	D	T_0	ADD	D	T_0	ADD	D	T_0	ADD	
Egg incubation period	9.68	10.5	**43.6**	5.21	10.5	**75.6**	2.53	10.5	**49.4**	**56.2**
Larval stage	30.08	8.5	**195.5**	14.32	8.5	**236.3**	9.40	8.5	**202.1**	**211.3**
Pupal stage	10.42	5.3	**101.1**	7.08	5.3	**139.5**	4.19	5.3	**103.5**	**114.7**
Total developmental time			**340.2**			**451.4**			**355.0**	**382.2**
Preoviposition period	10.60	5.2	**103.9**	5.53	5.2	**109.5**	4.20	5.2	**104.2**	**105.9**
Generation			**444.1**			**560.9**			**459.2**	**488.1**
Oviposition period	38.75	5.4	**372.0**	29.68	5.4	**581.7**	22.19	5.4	**545.9**	**499.9**
Postoviposition period	16.86	6.9	**136.6**	6.23	6.9	**112.8**	5.76	6.9	**133.1**	**127.5**
Adult female longevity	66.21	0.8	**940.2**	41.44	0.8	**1028.8**	32.15	0.8	**938.8**	**969.3**
Adult male longevity	45.96	1.3	**629.7**	28.57	1.3	**677.1**	23.92	1.3	**686.5**	**664.4**

Awadalla, Hagar (2010) reported that the total developmental times of the immature stages of *R. cardinalis* were 62.30, 41.10, 26.85, 18.85 and 12.7days when reared on *I. purchasi* at 16, 20, 24, 28 and 32 ˚C, respectively. Also, Abdel-Salam et al. (2013) recorded that the larval developmental time average lasted 29.65± 0.95 days when reared on *I. purchasi* nymphs and reread on 16 ˚C and 60± 5% RH.

Larval Feeding Capacity:

Data tabulated in Table (3) and Fig. (1) showed that food consumption of larval instars which reared on the third nymphal instar of *I. purchasi* at three constant temperatures 15, 25 and 30 ˚C ±1.

The average of total nymphal instar of *I. purchasi* which consumed during the four larval instars of *R. cardinalis* when it reared at 15˚C were 8.96 ± 0.41, 9.20 ± 0.44, 16.8 ± 0.51 and 22.05 ± 0.78 individuals, respectively. But when it reread at 25 ˚C were 12.17 ± 0.84, 15.91 ± 1.2, 20.98 ± 2.31 and 57.43 ± 2.96 individuals, consecutively. At 30 ˚C the feeding capacity of four larval instars were 10.59 ± 1.1, 12.96 ± 1.3, 17.89 ± 1.53 and 43.80 ± 1.79 individuals, successively. The total average of nymphal consumed during the larval stage of *R. cardinalis* when reared on third nymphal instar of *I. purchasi* were 57.01 ± 1.1, 106.49 ± 4.2 and 85.24 ± 3.4 individuals under three constant temperatures 15, 25 and 30 ˚C ±1, respectively.

Table (3): Influence of three constant temperature degrees on the predaceous efficiency of *Rodolia cardinalis* (Mulsant) larval instars and adult reared on third nymphal instar of *I. purchasi*

Stage	15 ˚C	25 ˚C	30 ˚C
Larval stage			
1st larval instar	8.96 ± 0.41	12.17 ± 0.84	10.59 ± 1.1
2nd larval instar	9.20 ± 0.44	15.91 ± 1.2	12.96 ± 1.3
3rd larval instar	16.8 ± 0.51	20.98 ± 2.31	17.89 ± 1.53
4th larval instar	22.05 ± 0.78	57.43 ± 2.96	43.80 ± 1.79
Total	**57.01 ± 1.1**	**106.49 ± 4.2**	**85.24 ± 3.4**
Adult			
Female	384.96	351.72	296.81
Fecundity	162	443.71	315.50
Male	203.4	197.81	175.45

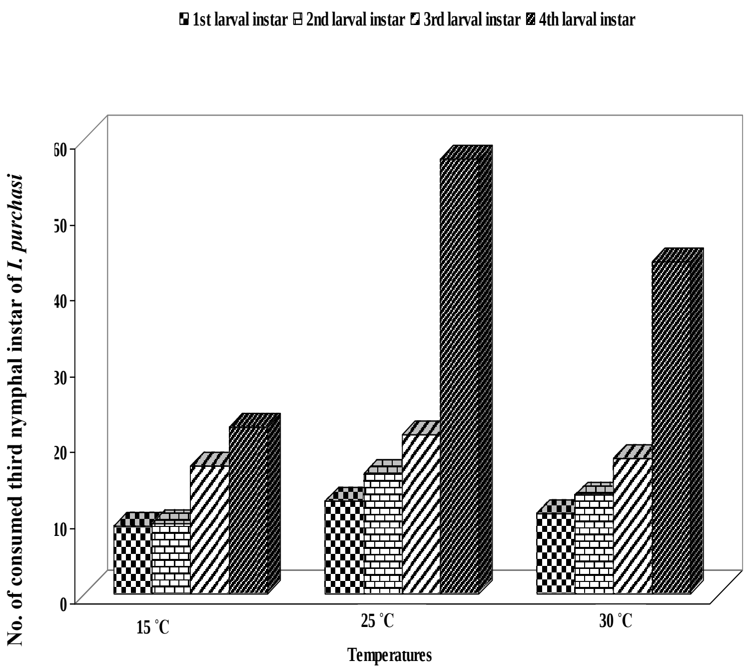

Fig. (1): Mean larval feeding capacity of *Rodolia cardinalis* (Mulsant) reared on third nymphal instar of *Icerya purchasi* at different temperatures

Pupal Stage:

Data arranged in Table (1) revealed that pupal developmental time lasted in 10.42, 7.08 and 4.19 days at15, 25 and 30 ˚C. Data tabulated in Table (2) showed that the lower developmental threshold of the predator was 5.3 ˚C and the thermal constant for their development was 114.7 DD unite.

These results were consistent with the observation of Grafton-Cardwell *et al.* (2005) who reported that development of the prepupal and pupal stages of *R. cardinalis* declined from 14.4 and 19.8 days at 14 ˚C to 1.7 and 4.5 days at 26 ˚C when they reared on *I. purchasi* nymphs.

Total Developmental Time:

The obtained results presented in Table (1) showed that the total developmental time of *R. cardinalis* was highly significant shortest (16.12 days) at 30 ˚C and highly significantly longest at 15 ˚C where 50.18 days when reared on *I. purchasi* nymphs.

This result was confirmed by the findings of Grafton-Cardwell *et al.* (2005) who reported that the total the developmental time from egg to adult emergence decreased from 79 to 18 days for temperatures from 14 to 25 ˚C.

Generation:

Data tabulated in Table (1) showed that the generation average of *R. cardinalis* was highly significant shortest (20.33 days) at 30˚C and highly significantly longest at 15 ˚C where 60.78 days when it reared on *I. purchasi* nymphs.

Data presented in Table (2) revealed that the average of accumulated degree days unite was 488.1 DD unite. These results were consistent with the observation of Grafton-Cardwell *et al.* (2005) who mentioned that the lower development thresholds differed according to life stage and ranged from 6.58 ˚C for the first instar to 13.39 °C for the prepupa. The smallest slopes (0.013 and 0.015) corresponded with the egg and pupal stages, which had the longest duration of development at all temperatures. Also, the development of vedalia from egg to adult eclosion required an average of 279 degree–days above a threshold of 10.8 ˚C.

Longevity:

Data presented in Table (1) showed that the average of life span for *R. cardinalis* adult female and male were decreased from 66.21 and 45.96 days to 23.15 and 23.92 days as the temperature increased from 15 to 30˚C, respectively.

Data tabulated in Table (2) indicated that the lower developmental threshold of *R. cardinalis* male and life span were 1.3 and 0.8˚C, successively. And the average of accumulated degree days unite for male and female longevity were 664.4 and 969.3 DD unite, consecutively.

Adult Male and Female Feeding Capacity:

Data obtained results presented in Table (3) and Fig. (2) cleared that through the adult life span, the feeding capacity of the adult female was 296.81 individuals at 30 ˚C and this value increased at 15˚C to reach maximum consumed which was 384.96 individuals. The average number of laid eggs per female (fecundity) was higher when the females were fed on *I. purchasi* nymphs at 25˚C with the value of 443.71 eggs/ female. While the low number of eggs was laid by the females that reared at 15 ˚C with the value of 162 eggs per female.

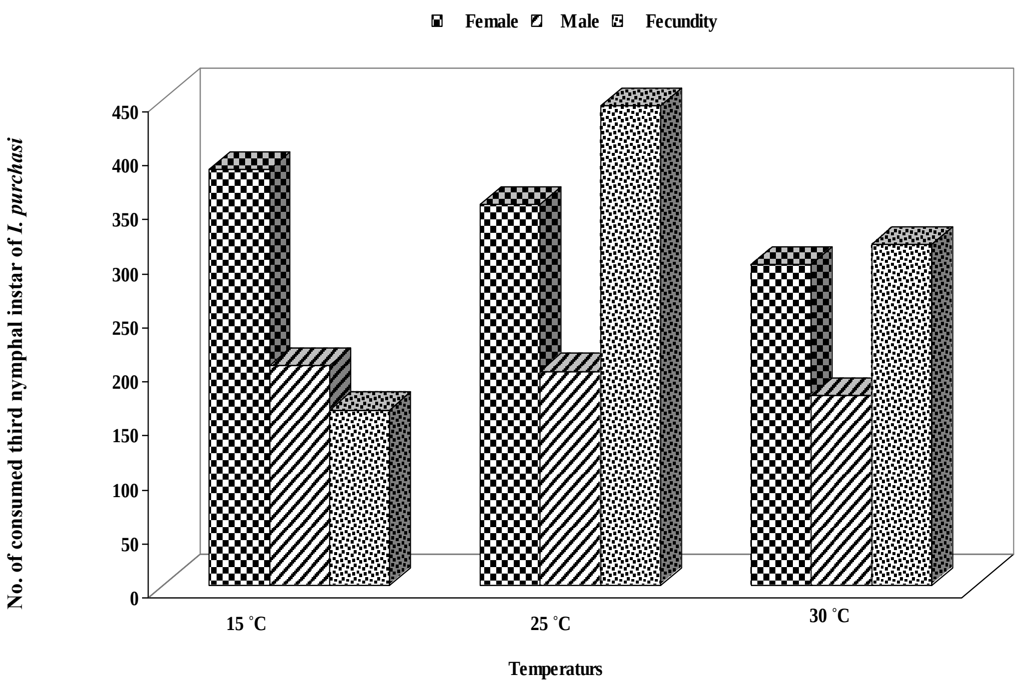

Fig. (1): Mean adult feeding capacity and female fecundity of *Rodolia cardinalis* (Mulsant) reared on third nymphal instar of *Icerya purchasi* at different temperatures.

The feeding range of *R. cardinalis* larvae is most likely defined when the adults select prey for oviposition. In our feeding tests and in the field (Quezada, 1969; Ragab, 1995), *R. cardinalis* laid eggs in or on its target prey, *I. purchasi*. This behavior suggests host specialization and has been observed in other species of coccinellids (Booth *et al.*, 1995; Kairo and Murphy, 1995; Lopez and Kairo, 2003). Although, oviposition was observed in some of the test arenas occupied by alternate prey, eggs were deposited haphazardly and beetles also laid eggs in the arenas with only water. This suggests that factors other than the presence of non target species were responsible for stimulating oviposition such as egg storage capacity in the oviduct (Dixon, 2000).

This is consistent with the study done by Hamed and Chemsedine (2001) who noticed that the fecundity of females at different temperatures ranged between 107 eggs at 15 ˚C and 601.86 eggs at 30 ˚C. The preoviposition period ranged between 23.75 days at 15 ˚C and 3.47 at 35 ˚C. Abdel-Salam (2013) found that the fecundity of *R. cardinalis*females was 336.6±25.43 eggs per female when reared *I. aegyptiaca* nymphs under the constant temperature of 28 and 70±5 R.H.

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ARABIC SUMMERY

العلاقة بين تطور الاطوار المختلفة للمفترس (Mulsant) *Rodalia cardinals* والمربى على *Icerya purchasi* Maskell والوحدات الحرارية المتجمعة

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١ – معهد بحوث وقاية النباتات- مركز البحوث الزراعية- الدقى- مصر
٢- قسم وقاية النبات- كلية الزراعة- جامعة المثنى- العراق

أُجريت التجارب المعملية في قسم الحشرات الاقتصادية بكلية الزراعة ، جامعة المنصورة لدراسة العلاقة بين تطور الاطوار المختلفة للمفترس(Mulsant) *Rodalia cardinals* والمربى على العمر الحورى الثالث للبق الدقيقى الاسترالى *Icerya purchasi* Maskell والوحدات الحرارية المتجمعة على ثلاث درجات حرارة ثابتة هى (١٥و ٢٥ و٣٠ درجة مئوية). أوضحت النتائج أن دورة حياة المفترس أطول عند درجة حرارة ١٥ درجة مئوية وبزيادة درجة الحرارة من ١٥ – ٣٠ م ° تقل طول فترة حياة الحشرة الكاملة للمفترس *R. cardinals* . وكان درجة حرارة صفر النمو لمراحل تطور المفترس المختلفة هى :١٠,٥ و٨,٥ و٥,٣ و ٥,٢ و٥,٤ و٦,٩ و ٠,٨ و ١,٣ درجة مئوية لكل من طور البيضة وطور اليرقة وطور العذراء وفترة ماقبل وضع البيض وفترة وضع البيض وفترة ما بعد وضع البيض وطول عمر الاناث وطول عمر الذكور على الترتيب.

وأظهرت النتائج أن الوحدات الحرارية المتجمعة اللازمة لمدة طور البيضة وطور اليرقة وطور العذراء وطول عمر الاناث وطول عمر الذكور هى : ٥٦,٢ و ٢١١,٣ و١١٤,٧ و ٩٦٩,٣ و ٦٦٤,٤ وحدة على التوالى.

أوضحت النتائج أن فترة حياة الأطوار الكاملة انخفضت بإرتفاع أو زيادة درجات الحرارة تدريجيا. وكان أعلى معدل للاستهلاك لكل أنثى على درجة حرارة ١٥ درجة مئوية مقارنة بدرجات الحرارة الاخرى. و لقد وضعت اناث هذا المفترس أعلى كمية من البيض حيث وصلت إلى ٤٤٣,٧١ بيضة/ أنثى على درجة حرارة ٢٥ درجة مئوية. وتتضح من النتائج المتحصل عليها امكانية التنبؤ بتعداد المفترس *R. cardinals* كذلك وقت اطلاق المفترس واستخدامه فى برامج المكافحة المتكاملة بالحقول المصرية وذلك لمكافحة البق الدقيقى الاسترالى.