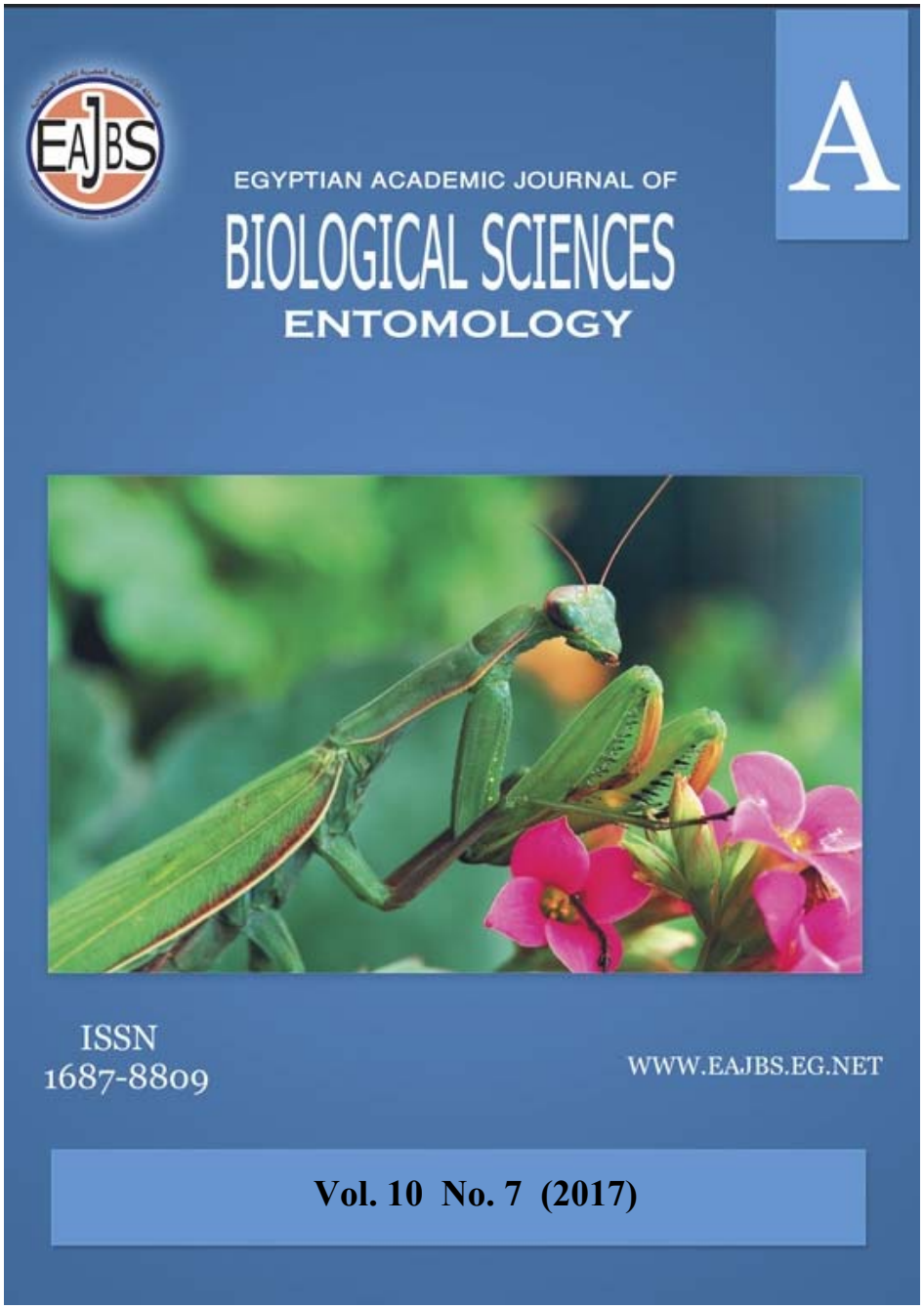

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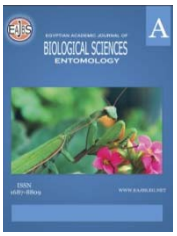

Studies on Some Parasitoids of Aphid *Aphis gossypii* Glover, (Homoptera: Aphididae) on Cucumber Plants in Egypt

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ABSTRACT

The present study was conducted to survey and population density of aphid *Aphis gossypii* Glover (Homoptera: Aphididae) infesting cucumber plants and its parasitoids were investigated at Diarb Nigem district during two season 2015 and 2016. Three primary parasitoids *Lysiphlebus fabarum* (Marshall), *Diaeretiella rapae* (M Intosh) and *Binodoxys angelica* (Haliday) and a hyperparasitoid, *Pachyneuron* sp. ((Hymenoptera: Aphidiidae)), were recorded. Primary parasitoid *Lysiphlebus fabarum* was the highly dominant species by relative density 53.76 and 52.24%, followed by *D. rapae* 22.58 and 27.73%, *Binodoxys angelica* 15.06 and 12.14%, while the hyperparasitoid , *Pachyneuron* sp., occurred by 8.60 and 7.89%, during 2015 and 2016 seasons respectively. The percentage of parasitism ranged from 3.14 % to 21.0 % in 2015, while it is starting by 2.66 % to reach its maximum 42.66% in 2016. Total developmental period of the parasitoid *Lysiphlebus fabarum* was 14.67 ± 1.16 days at 18.30°C ±1°C and 64 ±2RH%. Investigation the behaviour of the same parasitoid *Lysiphlebus fabarum* at varying host densities showed increase number of sting and number of mummies increased with increase of host density but decrease leaf arrival times and host arrival times with increasing . The obtained results showed that the longevity of the parasitoid *Lysiphlebus fabarum* was affected by temperature and food supply.

INTRODUCTION

Cucumber is the most important cucurbitaceous vegetable cultivated in Egypt in both the open field and under plastic houses. It cultivated in new reclaimed land for local consumption and exportation El-Lakwoh 2011. Cucurbitaceous vegetable plants are subjected to attack by numerous insect pests through the growing season such as , aphid , thrips and whitefly (El-Maghraby, *et al*. 1989 and El-Lakwoh 2011)

In addition, aphid have an important role in virus transmission (Waziery, 1996 and Barakat, 2005). *Aphis gossypii* (Homoptera: Aphididae) is the most common insect pests on cucumber (El-Maghraby *et al*., 1989 and Bennison, 1992).

The hymenopteran parasitoid, *Aphidius matricariae, Ephedrus cerasicola, Lysiphlebus testacipes, A. colemani* and *Trioxys auctus* are well known as potential bio agent for *A. gossypii* on cucumber (Steenis,1995; Albert, 1995; Hafez *et al.,* 1996 and Ohata, 2003).

The scope of the present study was directed towards the following.

1. Survey and population density of parasitoids associated with *A. gossypii* aphid and estimate the rates of parasitism in two seasons.
2. Estimating the effect of certain weather factors (Temperature and relative humidity) on the population density of the parasitoids and *A. gossypii* .
3. Biological aspects of parasitoid *Lysiphlebus fabarum* Such as, life cycle, behaviour the parasitoid on different host densities and Effect temperature and food supply on longevity.

MATERIALS AND METHODS

The present investigation was carried out at plant protection Research Institute, Sharkia Branch, Agricultural Research Center, and fields of (Diarb Negim) district Sharkia Governorate during 2015 and 2016 seasons.

Ecology:

Survey of the common parasitoids species associated with *A. gossypii* aphid on cucumber plant at Diarb-Negim district:

To survey the aphid parasitoids, random samples of the following aphid *Aphis gosspii,* which collected from cucumber plants cultivated at the experimental farm during the two seasons 2015 and 2016. The host plant was Cucumber, which kept free from any pesticide application .Weekly samples were chosen to be heavily infested plant parts with the previous aphid collected. Infested plant parts were placed in tight closed paper bags and transferred to the laboratory. The size of samples was five leaves of cucumber. All individuals from each aphid species found on the host plant samples were counted. Aphids were fed on their natural host and kept, every 50 aphids in a Petri dish until formation of mummies. The mummies of the aphids was isolated and separated in small glass tubes until the emergence of adult parasitoids. The emerged parasitoids were primarily classified, counted and preserved in 70 % ethyl alcohol. Parasitoid specimens were mounting, confirmation and identification was completed by aid of Prof. Ahmed El-Heneidy, biological control laboratory, Ministry of Agriculture, Egypt. The rates of parasitism caused by different parasitoid were calculated. In the laboratory, aphids were divided into 3 groups ; a- aphid mummies, b- living aphids containing parasitoid larvae (those were kept until the formation of mummies) and c- un parasitized aphids were recorded. The percentage of parasitism was calculated according to Farrell and Stufkens (1990).

Study the effect of certain weather factors on the population density of the aphid parasitoids cucumber:

To study the effect of certain weather conditions on the population density of the aphid parasitoids, the daily minimum, maximum and mean temperature and mean relative humidity were obtained from the Meteorological station, at Zagazig region. The correlation coefficient between weather parameters and the number of aphid parasitoids. Also, the numerical relation among these variables was calculated for the key weather factors, using regression coefficient (Fisher 1950)

Biology:

Life cycle of the parasitoid *Lysiphlebus fabarum* on *Aphis gossypii:*

A laboratory culture of the aphid *A. gosspii* was maintained under laboratory conditions. The aphid was reared on caged young seedling of its host (cucumber seeding) or on detached young leaves set flat on the bottom of clear plastic jar. The jar inverted so that the aphids fed in a natural position on under surface of the leaf

and change the leaf daily. A laboratory culture of the parasitoid *L. fabarum* started with mummies obtained from the field. Mummified aphids were placed singly in small glass tubes until the emergence of adult parasitoids which were fed on sugar solution. To determine the durations of different immature stages of the parasitoid *L. fabarum* on the nymphs of *A. gossypii*, nymphs were confined with the parasitoid for 5 hours. Forty nymphs of parasitized host aphid were daily dissected to determine the development of different immature stages of the parasitoid.

Effect of temperature and food supply on the adult longevity of the parasitoid *Lysiphlebus fabarum*:

Twenty mated females and twenty males of the parasitoid species were obtained from the laboratory culture (12hours) after adult emerged from aphid host *A. gossypii*. Each individual was confined in a small glass tube (9×2cm). The females and males of the parasitoid were divided into four groups, each of ten replicates, group (I) starved females and males, group (II) both sexes were supplied daily with 10% sugar solution and kept at room temperature (18.3°C) , group (III) adult female and males also starved and group (IV) supplied daily with 10% sugar solution but kept in refrigerator at (10°C) .

Adult stage behaviour of the parasitoid *Lysiphlebus fabarum* at varying host densities:

Lysiphlebus fabarum on varying host densities 40, 60, 80 and 100 nymphs of the aphid *A. gossypii* (mostly 3rd instars) on leaves of cucumber plant, were placed separately in Petri – dishes lined with moistened filter paper . Freshly emerged to 12 h. old molted parasitoid females, fully fed with 50% honey solution were gently introduced into each Petri-dish. The Petri dishes were covered with glass plates. The behaviour of the parasitoid was observed for 30 min and recorded, the period between introduction of the female and her first contact with the food plant , leaf (leaf-arrival time);and host (host – arrival time); and number of ovipositor (No. of stings)**.**

RESULTS AND DISCUSSION

Ecology:-

Survey of the common parasitoids species associated with *Aphis gossypii* aphid on cucumber plant at Diarb Negim district:

Aphis gossypii was the aphid species infesting cucumber crop. The primary parasitoids emerged from the mummified aphid were *Lysiphlebus fabarum*, *D. rapae*, *Binodoxys angelica*. Also, one secondary parasitoids: *Pachyneuron* sp. emerged from the mummified aphid. There results agree with Steenis (1995) who found that the parasitoid*, Ephedrus cerasicold, Lysiphlebus testaceipes* and *A. colemaili* 8 emerged from mummified aphid *A. gossypii* on cucumber. Meanwhile, Albert (1995) in German mentioned that *A. matricariae* parasitoid on aphid *A. gossypii* on cucumber plant. However, Hafez *et al*. (1996) in Egypt, studied the seasonal fluctuation of *A. gossypii* and associated predators and parasitoids Chrysopid species were the most abundant predators and *Trioxy auctus* first record on *A.gossypii* in Egypt. On the other hand, Ohta, (2003) showed that host acceptance and host suitability of the cotton aphid *A.gossypii* for *Lysiphlebus japonicas* and *A. colemani*.

Population density of *Aphis gossypii* and its common parasitoids on cucumber plant:

As shown in Figs. (1 and 2), four peaks for *A. gossypii* were recorded on cucumber plant in the first season 2015. These peaks occurred in the third week of

April (446 individuals) in the last week of April (475 individuals); in the second week of May (407 individuals) and in the fourth week of May (374 individuals).

Fig. (1). Three peaks for *A. gossypii* were recorded on cucumber plant in the second season 2016. These peaks occurred in the first week of April (601 individuals), during the first week of May (506 individuals) and during the first week of June (321individuals). Fig. (2). Fig. (1) Show that the maximum number mummified aphids in the first season 2015 was recorded in the fourth week of April (84 individuals) when the temperature and relative humidity were 18.3°C and 46.43%. While, the maximum number mummified aphids in the second season 2016 was recorded in the fourth week of April (183 individuals) when the temperature and relative humidity were 22.96°C and 38.71% R.H. Fig. (2).

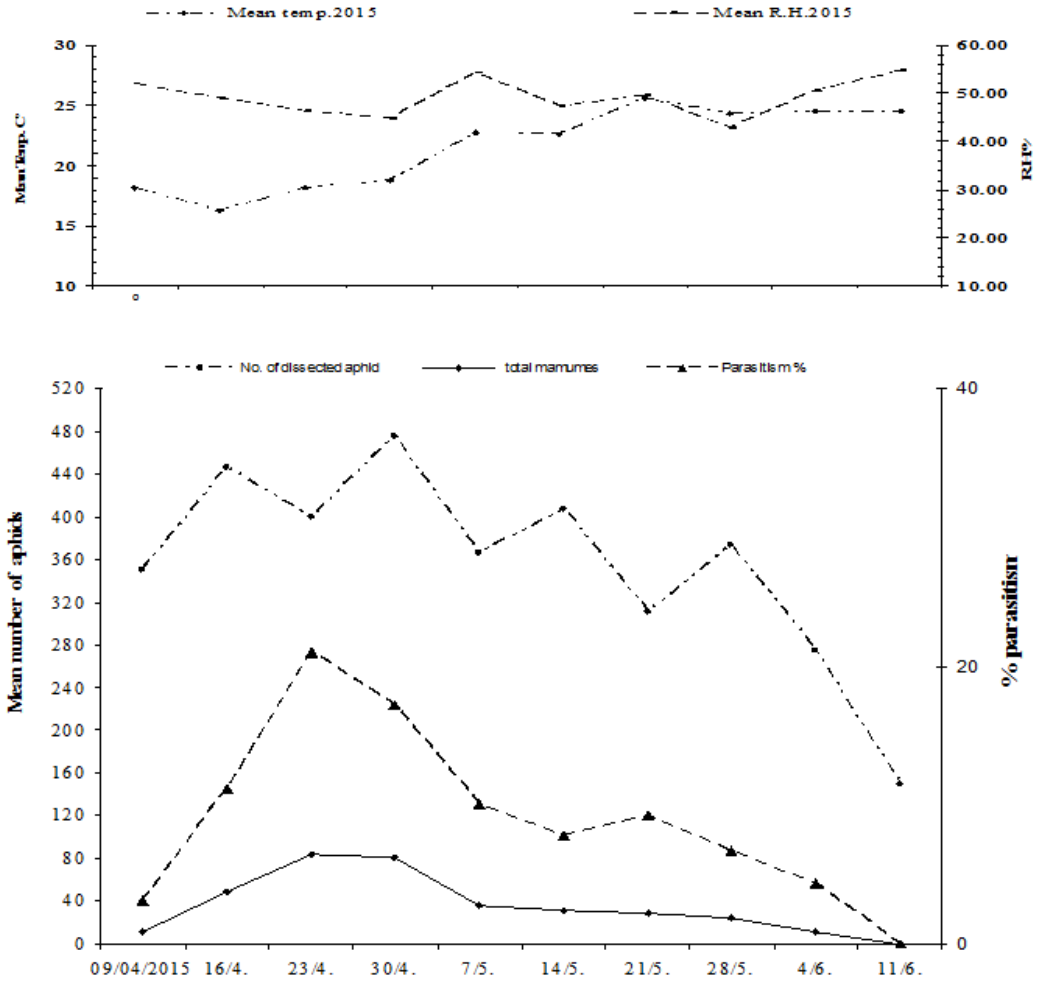


Fig. 1: Population density of *Aphis gossypii* aphid, number of parasitoid aphids and percentage of parasitism in cucumber field during 2015Season at Diarb Nagem district.

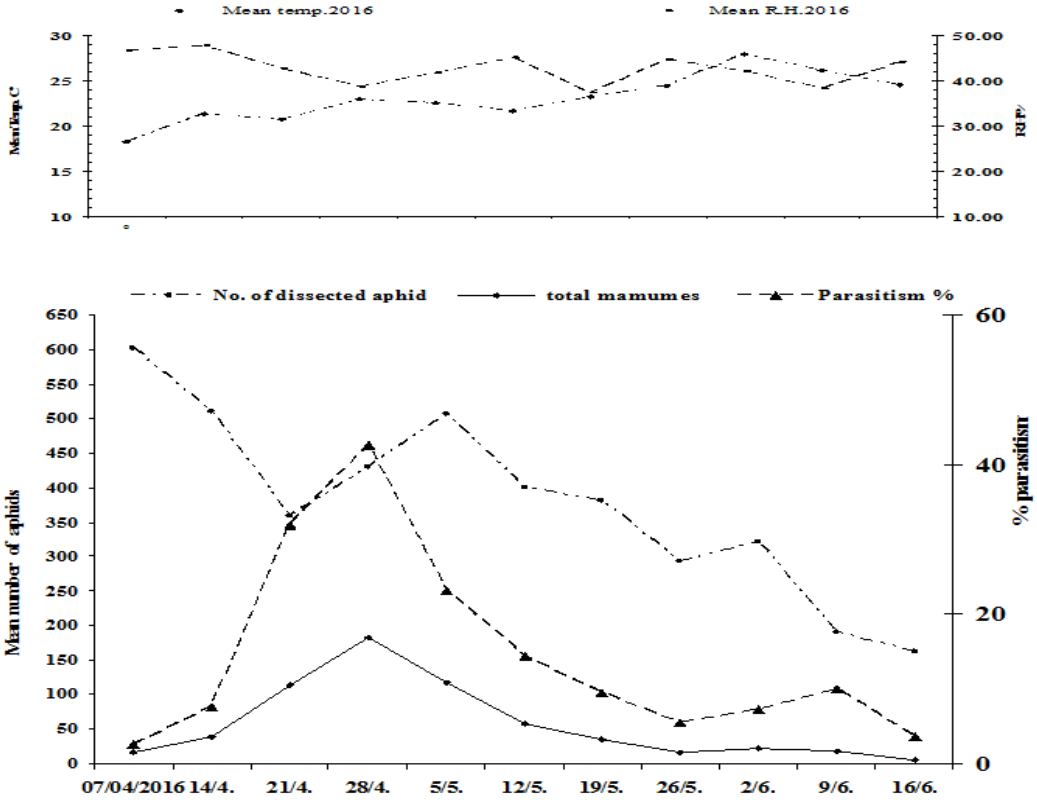


Fig. 2: Population density of *Aphis gossypii* aphid, number of parasitoid aphids and percentage of parasitism in cucumber field during 2016 season at Diarb Nagem district.

The data also cleared that *Lysiphlebus fabarum* a parasitoid was the most dominate species with high relative densities during the two successive seasons. In the first season 2015.*Lysiphlebus fabarum* adults began to appear on second week of April (8 individuals) then the number of parasitoid in increased gradually to reach the maximum (39 individuals) in the last week of April when the temperature and relative humidity were 18.71°C, 44.86% R.H. Fig. (3), while it had one peak in the second season 2016. This peaks occurred in the fourth week of April (85 individuals) when the temperature and relative humidity were 22.96°C, 38.71% R.H. Fig. (4).

The data also cleared that *D. rapae* a parasitoid for *A. gossypii* on cucumber had two peaks in the first season 2015. These peaks occurred in the fourth week of April (17 individuals), in the second week of May (5 individuals) when the temperature and relative humidity were 18.13°C, 46.43% R.H. and, 22.64°C, 47.43% R.H respectively (Fig. 3), while it had one peak in the second season 2016. This peaks occurred in the fourth week of April (39 individuals) when the temperature and relative humidity were 22.96°C, 38.71% R.H. Fig. (4).

The data also cleared that *Binodoxys angelica* a parasitoid for *A. gossypii* on cucumber had two peaks in the first season 2015. These peaks occurred in the fourth week of April (16 individuals), in the second week of May (5 individuals) when the temperature and relative humidity were 18.13°C, 46.43% R.H. and, 22.64°C, 47.43% R.H respectively Fig. (3), while it had one peak in the second season 2016. This peaks occurred in the fourth week of April (19 individuals) when the temperature and relative humidity were 22.96°C, 38.71% R.H. Fig. (4).

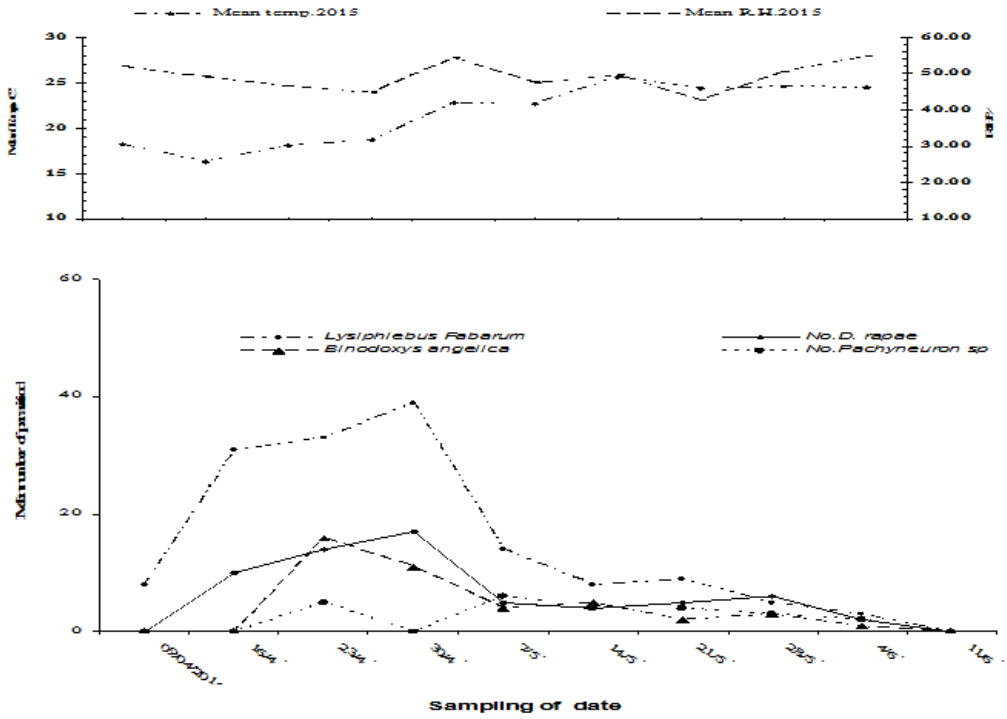


Fig. 3: Population density of *Aphis gossypii* aphid and number of parasitoid aphids in cucumber field during 2015season at Diarb Nagem district.

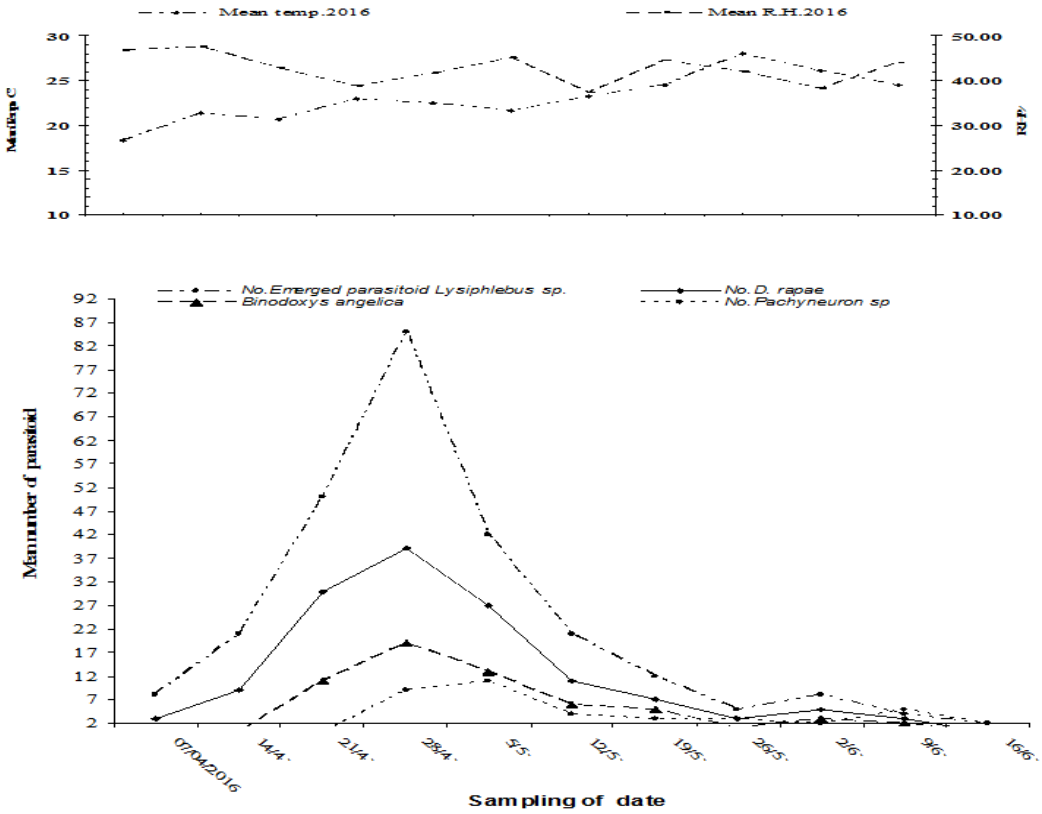
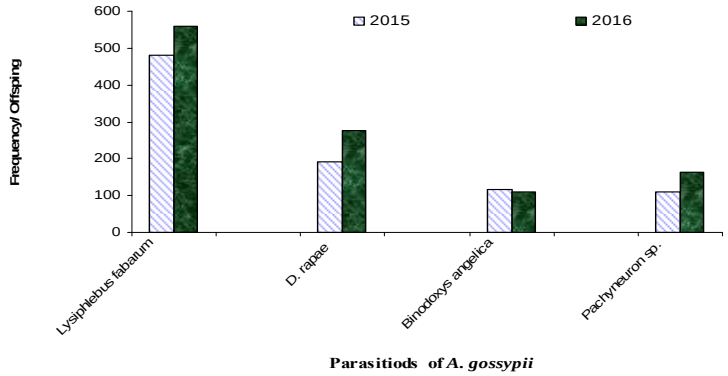


Fig. 4: Population density of *Aphis gossypii* aphid and number of parasitoid aphids in cucumber field during 2015\2016 season at Diarb Nagem district.

The data also cleared that *Pachyleuron* sp. a hyper parasitoid had few number of *Pachyneuron* sp . Adult was recorded during the two seasons. *Pachyneuron* sp was recorded during the period of April - June in the two *seasons* Figs. (3 and 4). Fig. (5). Shows the abundance percentage of *L .fabarum, D. rapae.* and *Binodoxys angelica and Pachyneuron sp.,* to the total catch of these parasitoids during the two seasons of study. The percentages were 53.76%, 22.58%, 15.06%and 8.60% in the first season 2015 and 52.24%, 27.73%, 12.14% and 7.89% in the second season



Percentage of parasitism:

The percentages of parasitism ranged from 3.14%to 21.0% in the first season 2015 (Fig. 1); while the percentages of parasitism starting by 2.66% in the second week of April and it increased until reached the peak of 42.66% in the second season 2016 (Fig. 2). The highest total parasitism ratio was 21.0% during the fourth week of April in the first season 2015 at 18.13°C and 46.43% R.H. as shown in Fig. (1), While it was 42.66% during the fourth week of April in the second season 2016 at 22.96°C and 38.71 % R.H. as shown in Fig. (2). The parasitism and development of *L. testaceipes on A.gossypii* was studied by Carnevale *et al.* (2003) on sweet pepper. Percentage of was higher 44.2% adult emergence 92.6%. However, the development and evaluation of an open rearing system for the control of *A. gossypii* by *L. testaceipes* in greenhouse on sweet pepper plants was recorded by Rodrigues *et al*. (2001) in Brazil. The parasitism percentage of *A. gossypii* ranged from 5 to 13%. On the other hand, Steenis (1995) recorded that *L. testaceipes* and *Ephedrus cerasicola* parasitized 26% to 23 % of *A. gossypii, A. colemani* parasitized 72.80 %of aphid while *Aphidius matricariae* parasitized less than 6% of *A. gossypii*.

Effect of weather factors on the population density of aphid *A. gossypii* and their parasitoids at Diarb -Nigm.

On *L. fabarum* :The obtain results indicated that the minimum temperature and mean temperature cleared highly negative significant correlation on the population density of *L.fabarum* (r= -0.8321 and -0.7776)but the maximum temperature showed negative significant correlation (r=-0.653)in the first season 2015 Table (1).

On *D. rapae* Negative significant correlation coefficient between minimum temperature, minimum relative humidity and the population density of the parasitoid *D. Rapae* (r =-0.6494 and -0.7399) in the first season Table (1). On the other hand Saleh *et al.* (2009). Cleared that highly significant correlation between temperature and *D. rapae* during the two seasons.

On *Binodoxys angelica.* The minimum relative humidity cleared a negative significant correlation with population of *Binodoxys angelica* on cucumber during the tow season of study (r= -0.5982 and -0.5219). Also, the minimum temperature cleared a negative correlation (r= -0.6811) in the season 2016 Table (1).

Table 1: Simple correlation between aphid parasitoid climatic Factors on "Cucumber" during of 2015 and 2016 season at Diarb- Nigm district.

Variable	Insect Pests			
	2015			
Source	*L.fabarum*	*D. rapae*	*B. angelica.*	*Pachyneura* SP.
Min Temp	-0.8321**	-0.6494*	-0.494	0.198
Max Temp	-0.6534*	-0.409	-0.163	0.486
Mean temp	-0.7776**	-0.555	-0.352	0.348
Min R.H	-0.602	-0.7399*	-0.5982*	-0.333
Max R.H	-0.037	-0.24	0.053	0.317
Mean R.H	-0.413	-0.631	-0.514	-0.064
		2016		
Min Temp	-0.498	-0.592	-0.6811*	-0.48
Max Temp	0.211	0.276	0.437	0.443
Mean Temp	-0.28	-0.275	-0.125	0.205
Min R.H	-0.368	-0.447	-0.5219*	-0.442
Max R.H	0.242	0.26	0.105	-0.281
Mean R.H	-0.281	-0.301	-0.486	-0.528

*=Significant Ns= not Significant

Biology:

Life cycle of *Lysiphlebus fabarum* on *Aphis gossypii* :

The results given in Table (2) indicate that the parasitoid egg lasted 2-3 days with an average of 2.79 ± 0.26 days. The larval stage durated an average of 5.75 ± 0.64 days with arrange of 4- 7 days . The prepupal and pupal stage ranged from 5-6 days with an average 6.13 ± 0.34 days. The total developmental period lasted for 11-17 days, with an average of 14.67 ± 1.16 days, However, Harizanova and Ekbom (1997) showed that the total developmental time of *Aphidius colemani* was 13.9 days when reared on *A. gossypii*. On the other hand Abdel - Samad (1996) in Egypt, the incubation period of *L. fabarum* tasted for an average of 0.40± 0.03 day under laboratory conditions. However, Carnevale *et al*. (2003). They reported that development time of the parasitoid *Lysiphlebus* sp on *A. gossypii* (8.8 days) and longevity was 5.5 days. These results agree with those of Ali (2014) in Egypt who mentioned that the biology of the parasitoid, *Lysiphlebus fabarum* on *A. craccivora* was 2.82 days for period of egg, larval period was 5.09 days, while pupal stage recorded 5.06 days. The total developmental periods averaged 12.42, at20°C.

Table 2: Duration of various developmental stage of *Lysiphlebus fabarum* reared on *Aphis gossypii* (average temperature 18.3 ± 1°C and 64 ± 2 RH)

Stage		Duration in days	
		Range	Mean ± SE
Egg		2-3	2.79 ± 0.26[c]
Larval		4-7	5.75 ± 0.64[b]
Pupal		5-6	6.13 ± 0.34[b]
Total development period(egg-adult)		11-17	14.67 ± 1.16[a]
Longevity	♀	4-7	5.83 ± 1.45[b]
	♂	2-3	2.61 ±0.32 [c]

Means followed by the same letter in a column are not significantly different at 0.05% level of probability.

Effect temperature and food supply on the adult longevity of the parasitoid *Lysiphlebus fabarum*.

From data presented in Table (3), it could be, generally, observed that adult females lived for a longer period than males ,irrespective of host aphid species from which the adults emerged , the nutritive solution on which the adults were supplied, and also the temperature at which the adults were kept. It could be , also observed that keeping of emerged adults at low temperature (10°C) led the *L. fabarum* adults to survive for 3-4 times or more longer than longevities recorded for adults kept at 18.3 °C, form data in the same Table (3), it could be also observed that longevities of *L. fabarum* adults emerged from *A. gosspyii* mummies were the longest (3.06-20.53 days for males and 4.03-33.3 days for females). As for the effect of supplying honey droplets for feeding *L. fabarum* compared to the starved adults, show that among adults emerged from *A. gosspyii* mummies fed males lived at 18.3°C for 5.57 days and 10°C for 20.53 days , opposed to 3.2 and 7.3 days , respectively for the starved adults. Correspondent longevities for females were 7.12 and 33.34days for fed adults and 4.03 and 14.64 for starved adults. However, **Stary (1970)** reported that the adult life span of adult parasitoids is affected by many factors such as temperature, humidity, food, presence or absence of hosts, etc. On the other hand, the obtained results agree with those obtained by **(Ragab *et al.*, 2002 and Saleh (2008).** who mentioned that the longevity was affected by temperature and food supply of the parasitoid *D. rapae.*

Table 3: Effect temperature and food supply on the adult longevity of the parasitoid *Lysiphlebus fabarum*.

Group	Treatment	Temp. °C	Adult longevity (days)			
			Female		Male	
			Rang	Mean±SE	Range	Mean±SE
A	-	18.3	3-5	4.03±0.14[c]	2-3	3.064±0.11[c]
B	+	18.3	5-8	7.12±0.22[c]	4-6	5.57±0.21[b]
C	-	10	12-16	14.64±0.33[c]	6-8	7.3±0.12[d]
D	+	10	25-40	33.34±1.16[a]	15-23	20.53±0.62[b]

Means followed by the same letter in a column are not significantly different at 0.05% level of probability.

Adult stage behaviour of the parasitoid, *Lysiphlebus fabarum* at varying host densities:

As reported by Brown *et al.* (1970), the leaf- reaching is a measure of the attractiveness potency of the semi chemicals emitted by the food plants and the host insects. Data in Table (4) indicate that the leaf-arrival time decreased with increasing the host population density. While, the number of oviposition (No. of stings) and number of resultant aphid mummies increased with increasing the host density. Pinto *et al.* (2004) showed that the role of volatile stimuli in the host-searching behaviour of the two parasitoid species *L .testaceipes* and *A. calemani* in relation to the host *A. gossypii* on cucumber plants. The two parasitoids species

respond to stimuli from the host plants of *A. gossypii* in a similar way to parasitoids of aphid pests in other crops. The maximum host-arrival time was 8.04±0.86min. when the host density was 40 and it started to decrease as the host density increased which became the minimum 1.68±0.24 min. at host density of 100 individuals . Wickremasinghe and Van- Emden (1992) the found that *L. Fabarum* responded positive to the odour of the host plant, such response was greater either to odour of host aphids or honeydew or a combination of them. The strongest response was to a combination of the plant and aphids. The maximum leaf-arrival time was 8.22±0.27min. when the host density was 40 and it started to decrease as the host density increased which became the minimum 2.13±0.16 min. at host density of 100 individuals . The time of the first sting increased as the host population increased. It was minimum 22.08±1.09 min. at host density of 40 individuals, while its maximum value was 7.98±0.47 min. at host density of 100 individuals. The number of stings (oviposition) increased as the host population increase. The lowest value was 9.0±0.71 at population of 40 individuals and reached the maximum value 82.0±2.05 when the host population became 100 individuals Table (4). Also, the number of the formed mummies increased by increasing the population density. Its minimum value was 5.2±0.58 at host population of 40 individuals and increased gradually to become maximum (22.4±1.03) when the host population became 100 individuals. The increased number of antennal encounters, oviposition and number of mummies with increase of host density might be due to increased concentration of the kairomones excreted by the host aphids. These kairomones enhance the activity of the parasitoid, thus increasing its potentiality to locate more host individuals (Srivastava and Singh, 1988; Saleh, 2008 and Saleh *et al.*, 2009).

Table 4: Behaviour of the parasitoid *Lysiphlebus fabarum* on cucumber at different *Aphis gossypii* densities.

Host density	Leaf-arrival time (min.)	Host arrival time (min)	First sting time (min.)	No. of stings (oviposition)	No. of mummies
40	**8.04 ± 0.86^{a}**	**8.22±0.27 ^{a}**	**22.01±0.47^{a}**	**9.0±0.71^{c}**	**5.2±0.58^{c}**
60	**6.21±0.29^{b}**	**6.47±0.37^{b}**	**16.26±0.37^{c}**	**37.4±2.02^{b}**	**12.6±1.21^{b}**
80	**3.22±0.22^{c}**	**4.24±0.19^{c}**	**14.15±0.57^{c}**	**43.0±2.60^{b}**	**15.0±1.52^{b}**
100	**1.68±0.24^{d}**	**2.13±0.16^{d}**	**7.98±1.09^{d}**	**82 ±2.05^{a}**	**22.4±1.03^{a}**
$LSD_{0.05}$	**0.8227**	**0.7889**	**2.0653**	**6.2276**	**3.4051**

Means followed by the same letter in a column are not significantly different at 0.05% level of probability

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ARABIC SUMMARY

دراسات على بعض طفيليات من القطن على نباتات الخيار فى مصر

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تم حصر ودراسة الكثافة العددية لمن القطن *Aphis gossypii* Glover الذى يصيب نباتات الخيار وطفيلياتة الحشرية فى منطقة ديرب نجم - محافظة الشرقية خلال عامى الدراسة ٢٠١٥-٢٠١٦ . واظهرت الدراسة حصر ثلاث طفيليات أولية :-

Lysiphlebus fabarum, Diaeretiella rapae (M Intosh) and *Binodoxys angelica* ونوع واحد من الطفيليات الثانوية .*Pachyneuron* sp.

واظهرت النتائج ان الطفيل الاولى *Lysiphlebus fabarum* اكثرهم تواجد حيث سجل كثافة نسبية (٥٢.٢٤، ٥٣.٧٦ %) ثم الطفيل *Diaeretiella rapae* (٢٢.٥٨ ، ٢٧.٧٣ %) ثم *Binodoxys angelica* (١٥.٦ ، ١٢.١٤ %) بينما سجل الطفيل *Pachyneuron* sp (٨.٦٠، ٧.٩٨ %) خلال عامى الدراسة على التوالى.

وأوضحت النتائج ان نسبة التطفل تتراوح بين (٣.١٤-٢١%) و (٢.٦٦-٤٢.٦٦%) خلال عامى الدراسة ٢٠١٥و٢٠١٦ .

واظهرت النتائج ان دورة حياة الطفيل *Lysiphlebus fabarum* على درجة حرارة ١٨.٣ ±١م درجة مئوية هى ١٤.٦٧ ±١.١٦ يوم. وكذلك دراسة سلوك الطفيل *Lysiphlebus fabarum* على كثافات العائل وكذلك وقت اول وخزه مع زيادة كثافة العائل وعلى العكس يزداد عدد الوخزات وعدد المومياوات مع زيادة كثافة العائل.