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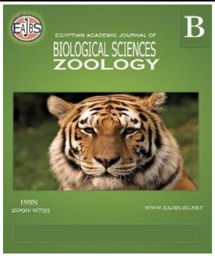

Estimating Seasonal Abundance of Piercing-Sucking Insects and Their Natural Enemies on Sweet Basil Plants

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ABSTRACT

Piercing-sucking insects are notorious devastating insects that cause substantial damage to crops and yield losses due to the direct feeding by both nymphs and adults furthermore through the transmission of viruses and diseases. Field experiments were conducted during the two growing summer seasons of 2021 and 2022 at Zagazig district, Sharkia Governorate, Egypt to assess the seasonal abundance of the major piercing-sucking insects and their associated natural enemies on sweet basil plants. Five piercing-sucking insect species were surveyed; *Bemisia tabaci* (Genn.), *Aphis gossypii* Glover, *Nezara viridula* L., *Empoasca decipiens* (Paoli), and *Thrips tabaci* Lind. Also, six predatory species were recorded; *Coccinella undecimpunctata* L., *Chrysoperla carnea* (Steph.), *Orius* sp., *Paederus alfierii* (Koch), *Metasyrphus corollae* (Fabr.) and *Scymnus* sp., and three parasitoid species were observed; *Aphidius colemani* (Viereck) as a primary parasitoid, *Alloxysta* sp. as a hyperparasitoid on *A. gossypii* and *Trissolcus megalocephalus* (Ashmead) as a parasitoid of *N. viridula* eggs. Our observations about the main piercing- insects and their natural enemies on sweet basil could be considered as a part of their control plan.

INTRODUCTION

Sweet basil (*Ocimum basilicum* L.) has been utilized as a medicinal and aromatic plant for a long time and its importance is still increasing globally, owing to its great economic value. (Mondal and Gupta 2016, Güez *et al*. 2017). Medicinal plants are not only used in the preparations of herbal medicine, but they are also used as food supplements, nutraceuticals, phytopesticides, and phytochemicals (Golan *et al*. 2019). Sweet basil is successfully cultivated in Egypt because of its outstanding adaptability to the Egyptian environment (Abuhashem *et al*. 2023).

Like other agricultural plants, sweet basil is vulnerable to being infected with various species of insects, causing substantial losses (Golan *et al*. 2019). Piercing-sucking insects are among the insect pests that exert noticeable damage to sweet basil plants (Ismail 2006, El-Ghanam 2017, Osman *et al*. 2017, Abou El-Saad 2018, Kumar *et al*. 2022). Whiteflies, cotton aphids, plant bugs, leafhoppers, and thrips are the most abundant and destructive insects on sweet basil (Sathe *et al*. 2014, Namvong and Chongrattanameteekul, 2015, Abou El-Saad 2018). They cause damage by direct feeding on the plant sap (Guerrieri and Digilio 2008, Kolaib *et al*. 2016) or by injecting toxic or by toxic salivary secretions during feeding and transmitting viral pathogens (Harrison *et al*. 1997, Saha *et al*. 2016). Furthermore, they secrete honeydew which may cause economic losses either by physical contamination or by providing a nutritional source for fungi growth and also disturb the photosynthesis process by blocking sunlight (Eid *et al*. 2018, Roonjho *et al*. 2022).

Hence, this study was carried out to estimate the seasonal abundance of piercing-sucking insect pests and their associated natural enemies on sweet basil plants.

MATERIALS AND METHODS

Field trials on sweet basil were carried out at Zagazig district, Sharkia Governorate, Egypt during the two growing seasons of 2021 and 2022. Sweet basil seedlings were transplanted in the second week of May in both seasons. Standard agronomic practices were performed as per requirement and no insecticides were applied throughout the period of the study.

Seasonal Abundance of the Main Piercing-Sucking Insect Pests and Their Predators:

At an area of 1000 m^2 (divided into three equal plots), the seasonal abundance of the major piercing-sucking insects and their associated predators were monitored weekly for 19 successive weeks, starting from the first week of June to the second week of October in the two consecutive seasons. Twenty leaves representing the three plant levels (upper, middle, and lower) were picked from each plot (following a random complete block design). The weekly samples were kept in paper bags and transferred to the laboratory for insect count using a stereoscopic microscope. The time of observations was between 6:00 to 8:00 a.m. Adults of injurious insects and their associated predators were evaluated by visual counts on the inspected plants with the aid of a (× 10) hand lens and the plants were carefully handled to avoid disturbance.

Seasonal Abundance and Parasitism Percentages of *A. gossypii* and *N. viridula* Parasitoids:

The total number of aphids per all samples (60 leaves of sweet basil) was determined weekly at random and divided into groups each of 50 aphid insects/Petri dish in the laboratory with fresh sweet basil leaves to identify and determine the percentages of aphid parasitoids. They were observed daily until the formation of mummies. The mummified aphids were counted and kept apart, segregated in Eppendorf tubes until the emergence of adult parasitoids. The emerged parasitoids were mostly counted to estimate the adult emergence percentages, then preserved in 70% ethyl alcohol until identification. The percentages of parasitism were estimated according to Farrell and Stufkens (1990):

$$Parasitism\ (\%) = \frac{the\ number\ of\ mummified\ aphids}{the\ number\ of\ aphids\ in\ all\ samples} x\ 100.$$

To evaluate the proportion of parasitism on green bug eggs, the leaves of sweet basil plants containing egg masses in all collected samples were gathered weekly and placed in Petri dishes (one egg mass/dish) under laboratory conditions (26±2°C and 65.00±5% R.H.). Each dish's bottom was covered with filter paper. Wet cotton was wrapped around the filter paper to moisten the leaf petiole. The egg masses were checked daily for the emergence of the egg parasitoids. The emerged adult parasitoids were collected and counted then preserved in 70% alcohol for identification. The number of parasitized eggs, percentage of parasitism, and sex ratio were determined. All parasitoid specimens were detected at the laboratory of Biological Control Department, Plant Protection Research Institute, Dokki, Giza, Egypt.

Statistical Analysis:

The differences between the numbers of piercing-sucking insects in the first and second seasons of the study, as well as the predators, were compared using the Paired T-Test with a probability of 5 and 1%. (SPSS 2015).

RESULTS AND DISCUSSION

Seasonal Abundance of the Main Piercing-Sucking Insect Pests and Their Predators:

Regarding the weekly inspections of sweet basil plants, it was observed that they were mostly liable to infestation with *Bemisia tabaci* (Genn.), *Aphis gossypii* Glover, *Nezara viridula* L., *Empoasca decipiens* (Paoli) and *Thrips tabaci* Lind. These results are consistent with those of Ismail (2006) who surveyed the main insect pests attacking 23 medicinal and aromatic plants including sweet basil in five Governorates in Egypt and recorded the whiteflies, cotton aphids, cotton thrips, jassids, and green bugs as major insect species on sweet basil plants. Moreover, Namvong and Chongrattanameteekul (2015) reported that *T. tabaci* was the most abundant insect species on sweet basil in Thailand.

The occurrence of *B. tabaci* was noticed on sweet basil plants throughout the two growing seasons. Three activity peaks were detected in the first season and two in the second one. The maximum number of *B. tabaci* in both seasons was recorded in the third week of July by 503 and 407 individuals/sample, alternatively. The initial incidence of the cotton aphid was detected from the first week of June then the insect population increased gradually forming two peaks, the first one was the most numerous recorded in the third week of July with 128 and 104 individuals/sample in 2021 and 2022 seasons, subsequently. The main period of activity for *N. viridula* on sweet basil plants mostly occurred in June, July, and August. During this period the insect numbers fluctuated and peaked in the first week of July (153 and 112 individuals/sample), in the two seasons, respectively. The population of *E. decipiens* showed one distinct activity peak in the second week of August giving numbers of 44 and 49 individuals/sample in the two investigation seasons, subsequently. *T. tabaci* was observed on sweet basil plants from the second week of June. Afterward, the insect numbers increased and attained their maximum in the third week of July with 25 and 28 individuals/sample during the two growing seasons, respectively (Figs. 1 & 2 C). These results are in partial agreement with those of Abou El-Saad (2018) who studied some ecological attributes of major pests on sweet basil in Assuit Governorate, Egypt, and mentioned that the maximum populations of *B. tabaci*, *E. decipiens*, and *T. tabaci* were achieved in July while *A. gossypii* reached its peak in August. In general, the populations of piercing-sucking insects were relatively low throughout the early season and increased progressively by the growth of the plants then they gradually decreased towards the final stage of the plant growth. These may be due to the availability of plant sap in leaves even during the flowering period. Afterward, the nutritive sap levels decreased in the leaves of the plants when the fruits began to form. (Eid *et al*. 2018).

Data also revealed that these aforementioned insects were accompanied by six predaceous species; *Coccinella undecimpunctata* L., *Chrysoperla carnea* (Steph.), *Orius* sp., *Paederus alfierii* (Koch.), *Metasyrphus corollae* (Fabr.) and *Scymnus* sp. The majority population of them was found during the period confined between the fourth week of June to the fifth week of August. The mean numbers of *C. undecimpunctata* reached their maximum (19 & 17 individuals/sample), in the second week of August and the fourth week of July during the two studied seasons. In the first season, *C. carnea* showed two equal peaks (15 individuals/sample) in the third and fourth weeks of July while in the second one, it peaked once by 14 individuals/sample. The highest population levels of *Orius* sp. (8 & 10 individuals/sample) were observed in the second week of August and in the third week of July in the 2021 and 2022 seasons, successfully. In the first season, the maximum peak of activity of *P. alfierii* (18 individuals/sample) was discovered in the second week of August. In the second season, it had two equal peaks (17&17 individuals/sample) in the first and second weeks of August. *M. corollae* and *Scymnus* sp. were presented by few numbers in both seasons. (Figs. 1&2 B). Such findings are confirmed by Osman *et al*. (2017) and Abou El-Saad (2018). Previous studies have reported an association between the presence of sweet basil plants and the colonization rates and population persistence of lacewing by assessing how sweet basil, as a functional plant, affects the lacewing under laboratory and greenhouse conditions and found that the presence of sweet basil enhanced the fitness attributes of lacewing as compared with control treatments (Fang *et al*. 2022). Furthermore, the attractiveness of sweet basil plants and another four floral plants to three predator species and found that *P. alfierii* showed its maximum preferability rate to sweet basil flowers (Abd El-Kareim *et al*. 2011).

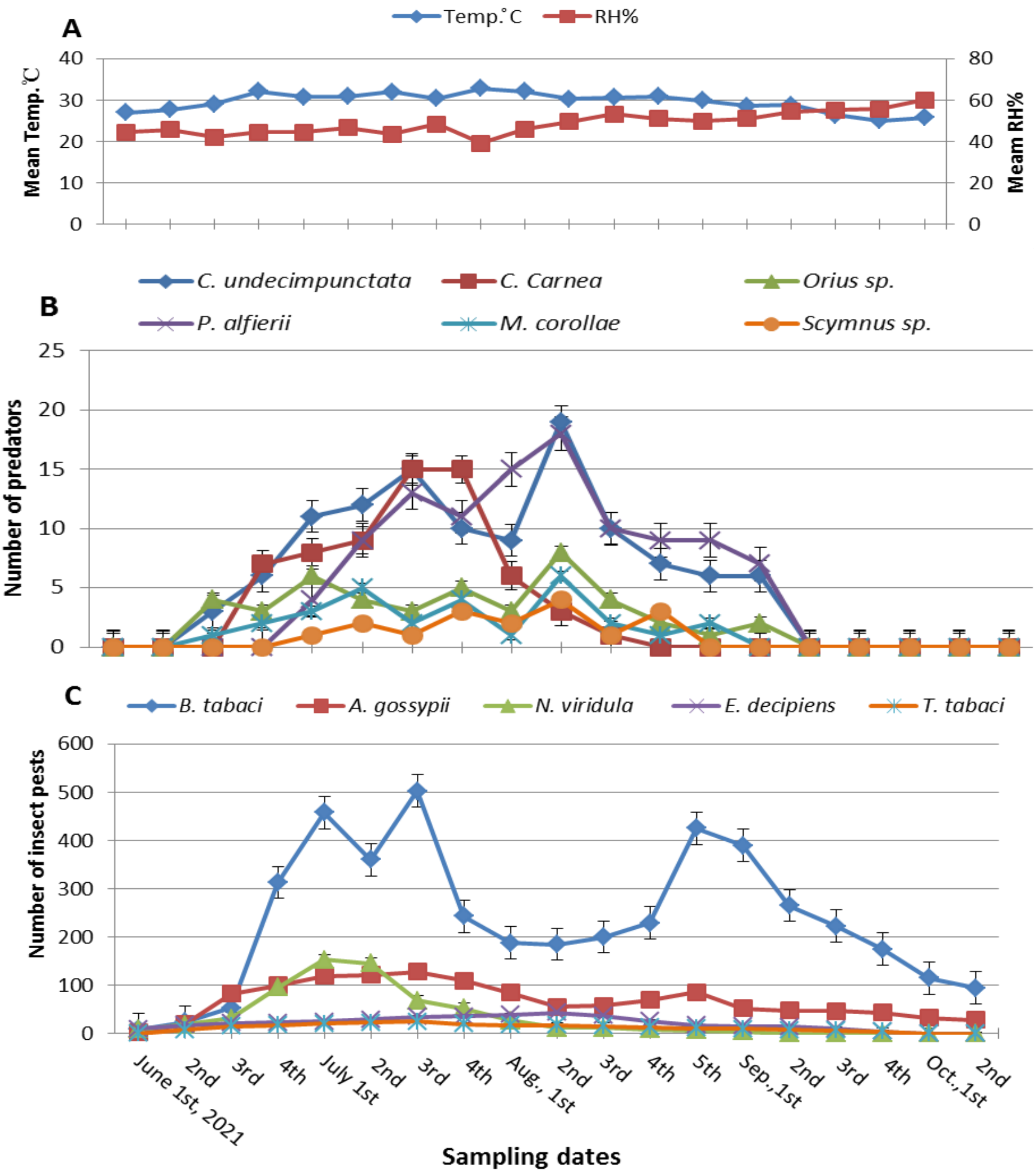

Fig. 1: Seasonal abundance of piercing-sucking insects infesting sweet basil plants **(C),** their associated predators **(B)** and weather factors **(A)** at Zagazig district, Sharkia Governorate, Egypt during the 2021 season.

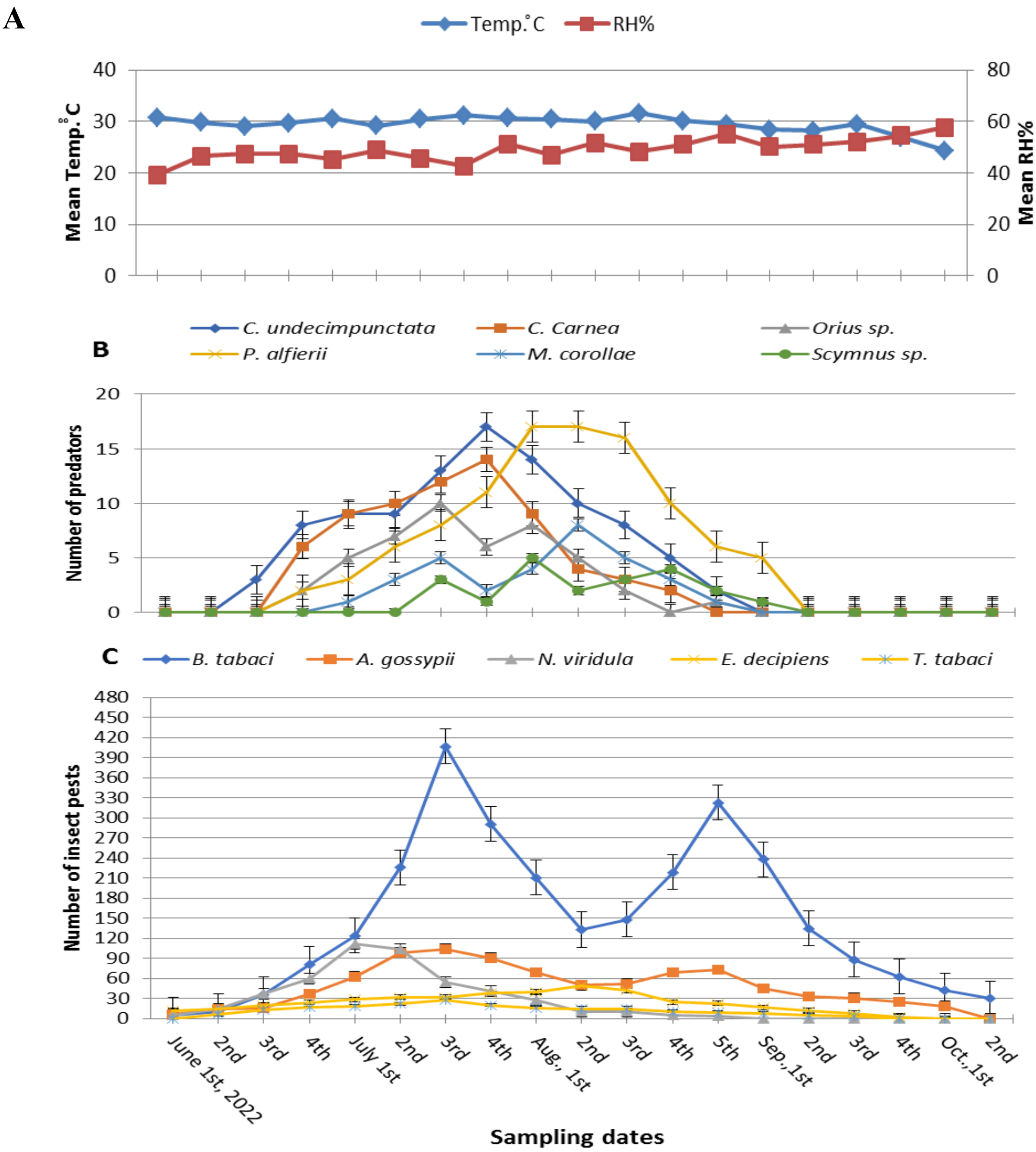

Fig. 2: Seasonal abundance of piercing-sucking insects infesting sweet basil plants**(C),** their associated predators **(B)** and weather factors **(A)** at Zagazig district, Sharkia Governorate, Egypt during the 2022 season.

The average means of temperature and relative humidity % ranged between 25.00 and 32.81°C, and 39.06 and 60.03% R.H. in 2021 and between 24.31 and 31.60°C, and 39.29 and 57.80% R.H. in 2022. It was noticed that the peaks of most piercing-sucking insects under study were much higher during the first season than the second one as there were slight decreases in the two weather factors throughout the second season. (Figs. 1 & 2 A).

Statistical analysis showed that there were highly significant differences between the insect population during the first and second study seasons, while the differences in the population of leafhoppers were significant, and in case of thrips, these differences were not significant at all. As for predatory insects, the differences between their numbers in both seasons were not significant with the exception of *C. undecimpunctata*, these differences were significant, (df = 56).

Seasonal Abundance and Parasitism Percentages of *A. gossypii* and *N. viridula* Parasitoids:

Two main hymenopterous parasitoid species emerged from the mummified aphid during the present investigation; a primary parasitoid (*Aphidius colemani* (Viereck)) and a hyperparasitoid species (*Alloxysta* sp.).

The main parasitism period of *A. colemani* was from the third week of June to the last week of August and the maximum number was detected in the fourth week of July. *Alloxysta* sp. appeared in a short term by a few numbers (from the first week of August to the second week of September). The percentage of parasitism started at 2.41% and increased gradually reaching its maximum (9.41%) in the first week of August then it decreased until the end of the first season. The existence period of *A. colemani* in the second season was noticed in July and August and the highest mean of numbers was recorded in the fourth week of August. *Alloxysta* sp. appeared rarely with limited numbers and the percentages of parasitism ranged from 1.59% to 10.14% (Figs. 3 & 4). Our results are supported by those of (Budenberg 1990) who stated that aphids may have evolved mechanisms to attract hyperparasitoids. Indeed, some secondary parasitoids are attracted by the volatiles from aphid honeydew. Aphid reproduction also increases in the presence of volatile chemicals released from hyperparasitoids without physical contact in the field, suggesting that aphids respond to chemical signals from hyperparasitoids (Boenisch *et al*. 1997, Van Veen *et al*. 2001). However, there is a need for more field experiments to investigate the multitrophic interactions within the aphid/parasitoid system over time and evaluate the possible consequences of pest control (Mkenda *et al*. 2019). In addition, previous studies identified plants including sweet basil as having positive effects on the parasitoid's longevity and parasitism rates (Charles and Paine 2016, Jado *et al*. 2019). Also, Souza *et al*. (2019) reported that sweet basil is an attractive and nutritious plant for parasitoids.

All the emerged parasitoids from the collected egg masses of *N. viridula* were identified as *Trissolcus megalocephalus* (Ashmead) (Hymenoptera: Scelionidae). The parasitism was first observed in the fourth week of June with a value of 8.16%, and it increased progressively to exhibit its highest rate (42.39%) in the third week of July in the first season of the study. In the second season, the first record of parasitism was in the third week of June, being 3.37%. In the second week of July, the range of parasitism increased reaching 51.75%. The general means of parasitism were 21.84% and 29.61% in the 2021 and 2022 seasons, successfully. The highest percentage of adult emergence for *T. megalocephalus* was 92.31% in the third week of July and the minimum was 71.43% in the first week of July in the 2021 season. In the 2022 season, the maximum emergence percentage of the parasitoid's adults was observed in the third week of July by 92.45% and the minimum percentage was 60.00% in the fourth week of June. The highest sex ratio (as females), 2 females: 1 male was achieved in the fourth week of June in both studied seasons. The mean sex ratio was 1.47♀:1♂ in the 2021 season and 1.40♀:1 ♂in the 2022 season (Table 1). The present results relatively agreed with those of Khalafalla *et al*. (2005) who reported that the highest rate of parasitism with *T. megalocephalus* on green bug eggs was recorded in July and August and it was synchronized with the increase in temperature.

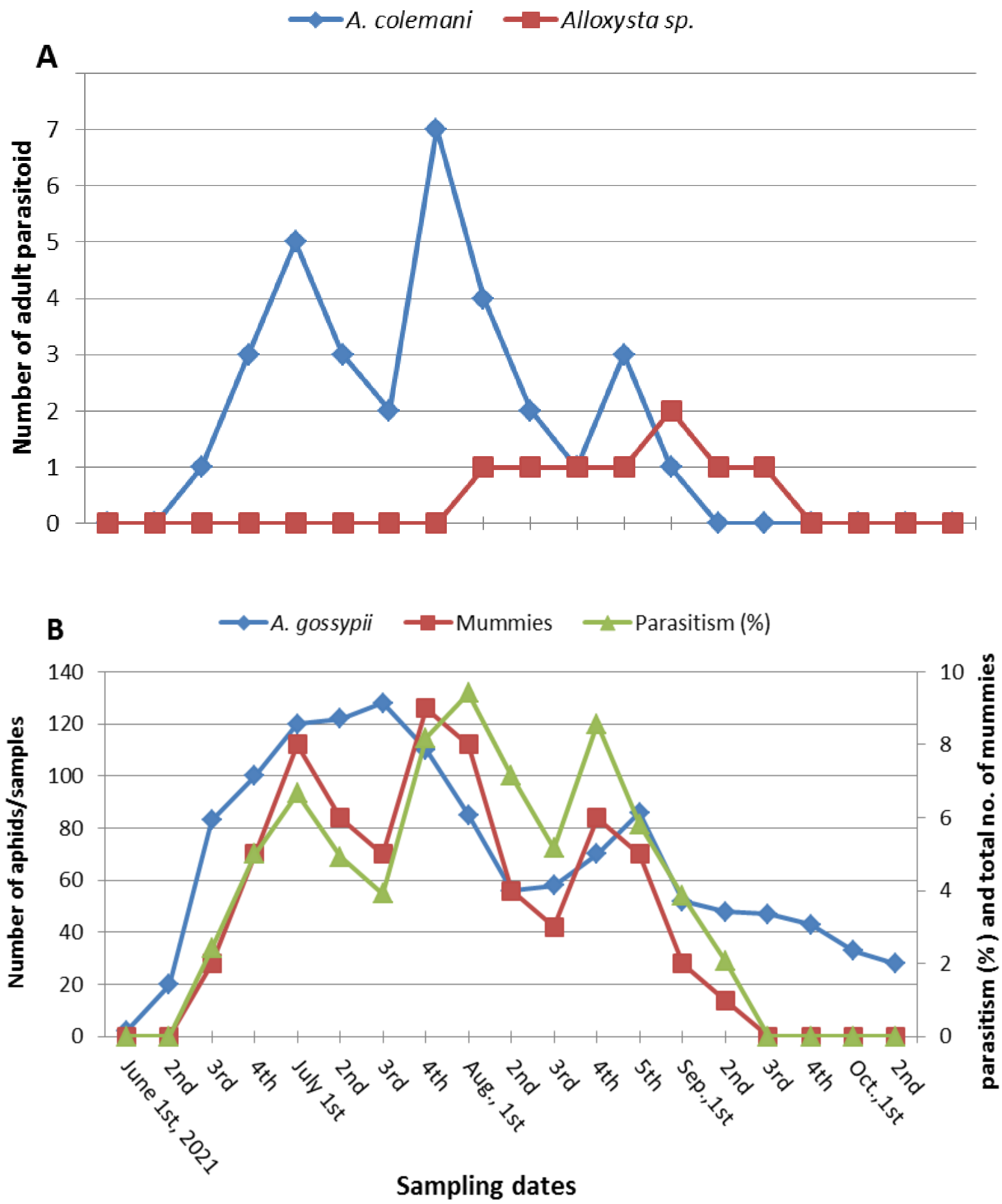

Fig.3: Seasonal abundance of primary parasitoid, *A. colemani* on *A. gossypii* and the hyperparasitoid, *Alloxysta* sp. (**A)** as well as Parasitism % and the total number of mummies (**B**) on sweet basil plants at Zagazig district, Sharkia Governorate, Egypt during 2021 season.

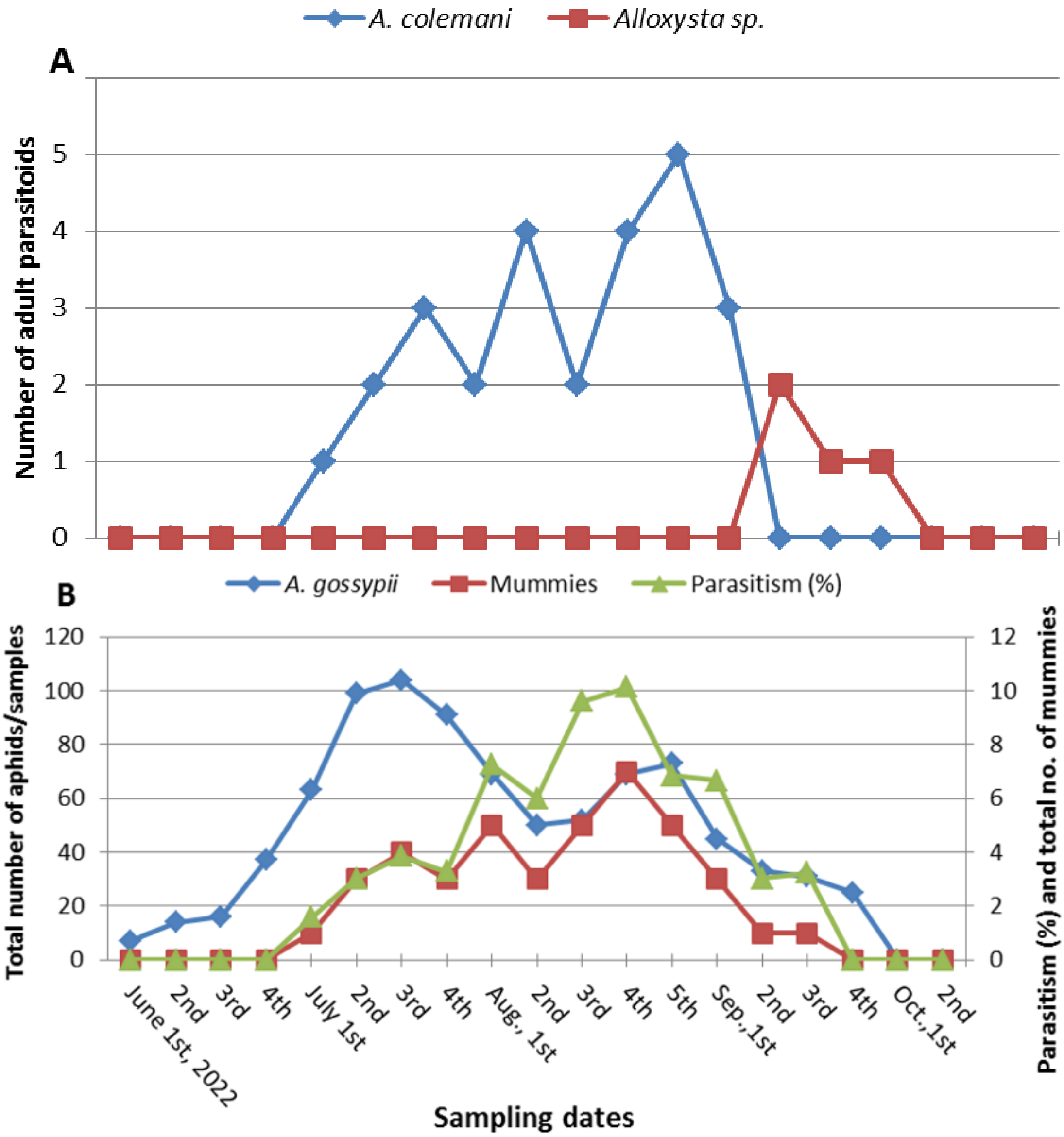

Fig. 4: Seasonal abundance of primary parasitoid, *A. colemani* on *A. gossypii* and the hyperparasitoid, *Alloxysta* sp. (**A**) as well as Parasitism % and the total number of mummies (**B**) on sweet basil plants at Zagazig district, Sharkia Governorate, Egypt during 2022 season.

Table 1: Seasonal abundance and parasitism percentages of *T. megalocephalus* on *N. viridula* eggs existing on sweet basil plants at Zagazig district, Sharkia Governorate, Egypt during the 2021 and 2022 seasons.

Sampling date	No. of egg masses		Total no. of eggs/masses		No. of Eggs Parasitized		Parasitism%		No. of adult emergence		Parasitoid adult emergence%		parasitoid Sex ratio (♀:♂)	
	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022
June, 2nd	00	00	00	00	00	00	00	00	00	00	00	00	00	00
3rd	00	2	00	89	00	3	00	3.37	00	2	00	66.67	00	1:1
4th	1	2	49	98	4	5	8.16	5.10	3	3	75.00	60.00	2:1	2:1
July, 1st	1	3	57	158	7	34	12.28	21.52	5	26	71.43	76.47	1.5:1	1.17:1
2nd	2	2	123	114	25	59	20.33	51.75	21	51	84.00	86.44	1.33:1	1.22:1
3rd	3	2	184	119	78	53	42.39	44.54	72	49	92.31	92.45	1.18:1	1.23:1
4th	2	1	119	47	31	20	26.05	42.55	28	18	90.32	90.0	1.33:1	1.57:1
Aug., 1st	00	1	00	39	00	15	00	38.46	00	13	00	86.67	00	1.6:1
2nd	00	00	00	00	00	00	00	00	00	00	00	00	00	00
Mean±SE	**1.8 ±0.37**	**1.86 ±0.26**	**106.4 ±24.68**	**94.86 ±15.72**	**29.00 ±13.28**	**27.0 ±8.45**	**21.84 ±6.00**	**29.61 ±7.42**	**25.8 ±12.47**	**23.14 ±7.61**	**82.61 ±4.11**	**79.81 ±4.70**	**1.47:1**	**1.4:1**

Conclusion

B. tabaci, *A. gossypii*, *N. viridula*, *E. decipiens*, and *T. tabaci* were the key piercing-sucking insect species on sweet basil plants, and they were accompanied by six predaceous species; *C. undecimpunctata*, *C. carnea*, *Orius* sp., *P. alfierii*, *M. corollae* and *Scymnus* sp. and three parasitoid species; *A. colemani*, *Alloxysta* sp.(as primary and hyperparasitoid on *A. gossypii*, respectively) and *T. megalocephalus* (as a parasitoid of *N. viridula* eggs). The existing predators and parasitoids with piercing-sucking insects in the field may help in suppressing these pests and decreasing the application of insecticides.

Declarations:

Ethical Approval:Ethical Approval is not applicable.

Conflicts of Interest: The authors claim that there are no conflicts of interest.

Authors Contributions:I hereby verify that all authors mentioned on the title page have made substantial contributions to the conception and design of the study, have thoroughly reviewed the manuscript, confirm the accuracy and authenticity of the data and its interpretation, and consent to its submission.

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Availability of Data and Materials: All datasets analysed and described during the present study are available from the corresponding author upon reasonable request.

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ARABIC SUMMARY

تقدير الوفره الموسميه للحشرات الثاقبه الماصه وأعدائها الطبيعيه على نباتات الريحان الحلو

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ينتج عن الاصابه بالحشرات الثاقبه الماصه فقد كبير في المحصول الناتج وذلك اما نتيجة التغذيه المباشره لكلا من الحوريات والحشرات الكامله أو بسبب النقل لعديد من الأمراض الفيروسيه. تم إجراء التجارب الحقليه خلال موسمي ٢٠٢١ و٢٠٢٢ في منطقة الزقازيق – محافظة الشرقيه –مصر لدراسة الوفره الموسميه للحشرات الثاقبه الماصه الرئيسيه والأعداء الطبيعيه المصاحبه لها على نباتات الريحان الحلو. وقد تم حصر خمسة أنواع من الحشرات الثاقبه الماصه؛ *B. tabaci* ،*A. gossypii* ،*N.viridula* ،*E.decipiens* و *T. tabaci* . كما تم تسجيل ستة أنواع من المفترسات *C.undecimpunctata* ،*C.carnea* ،*Orius* sp. ،*P.alfierii* ،*M.corollae* و *Scymnus* sp تم تسجيل ثلاثة أنواع من الطفيليات؛ *A. colemani* كطفيل أولي و *Alloxysta* sp. كطفيل مفرط على حشرة من القطن. وكذلك طفيل *T. megalocephalus* كطفيل على بيض البقه الخضراء. يمكن اعتبار هذه الملاحظات حول الحشرا ت الثاقبه الماصه الرئيسيه واعدائها الطبيعيه على نباتات الريحان جزء من خطة مكافحتها