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Population Abundance of *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillaridae) and its Parasitoids on some Citrus species in Surman region

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بسم الله الرحمن الرحيم

﴿ نرفع درجات من نشاء وفوق كل ذي علم عليم ﴾

صدق الله العظيم

سورة يوسف، الآية (76)

DEDICATION

I dedicate this effort to the dearest persons in my heart... (my parents) whom I pray to **Allah** to protect them

TO

The man whose name I proudly carry, **my father** Thank you so much, if it weren't for you, I wouldn't be where I am today

The woman who taught me patience and trust in Allah, **my mother** she helped me to overcome the odds and supported me in my weak Thank you very much my first and foremost teacher

my brothers and **sisters**...you have all my love….

my friend **Fatima** for supporting me….

everyone who helped me and saw that I deserved it….

for **everyone** who taught me a letter.

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I would like to extend my thanks to **Dr. Shaaban Amara** for making his farm available to conduct this study

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I would like to thank everyone who helped me directly or indirectly.

ABSTRACT

The present study aimed to investigate the population abundance of citrus leaf miner *Phyllocnistis Citrella* and the associated parasitoids on four host plants. 100 infected leaves were collected from each host plant weekly. Samples were kept in plastic bags and transferred for examination in the laboratory.

The results showed that *P. citrella* larvae recorded four peaks of abundance on Lemon occurred on the 21st of July, the 18th of August, the 17th of November, and the 1st of December, and five peaks of abundance on Washington navel occurred on the 14th of July, the 25th of August, 8th of September, 22nd of September and 6th of October respectively, while recorded five peaks of abundance on Tarocco, occurred on the 30th of June, the 21st of July, the 1st of September, the 15th of September, and the 10th of November, More over four peaks of abundance recorded on Hasna occurred on the 14th of July, the 11th of August, the 25th of August, and the 1st of September.

P. citrella pre pupa recorded four peaks of abundance on Lemon occurred on the 7th of July, the 25th of August, the 27th of October, and the 1st of December, and three peaks of abundance on Washington’s navel occurred on the 7th of July , 14th of July and 20th of October and recorded three peaks of abundance on Tarocco occurred on the 30th of June, the 7th of July, and the 20th of October, while recorded on Hasna three peaks of abundance occurred on the 7th of July, the 11th of September, and the 20th of October.

P. citrella pupa recorded five peaks of abundance on Lemon occurred on the 16th of June, 30th of June, 10th of January, 17th of January, and 8th of March, and recorded five peaks of abundance on Washington's navel occurred on the 22nd of May, 9th of June, 16th of June, 30th of June and 7th of July and recorded four peaks of abundance on Tarocco occurred on the 9th of June, the 30th of June, the 20th of October, and the 27th of October while recorded three Peaks of abundance on Hasna

occurred on the 30th of June, the 7th of July, and the 20th of October respectively.

The ectoparasitoid *Semielacher petiolatus* was determined to attack *P. citrella* which recorded four peaks of abundance on Lemon occurred on the 1st of September, 20th of October, 24th of November, and 1st of December and recorded four peaks of abundance on Washington navel occurred on the 7th of July, 14th of July, 1st of September, and 8th of September and recorded five peaks of abundance on Tarocco on the 14th of July, 4th of August, 18th of August, 25th of August, and 1st of September while recorded three peaks of abundance on Hasna occurred on the 11th of August, 1st of September, and 20th of October respectively.

Correlation analysis indicated a positive correlation between the average temperature and the population of *P. citrella* with an r-value (0.74) and (0.69) with *S. petiolatus* while correlation analysis showed a negative correlation and insignificantly between the relative humidity and the population of *P. citrella* with an r-value (- 0.22) and (- 0.11) with *S. petiolatus.*

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The summary	full name
P. citrella	*Phyllocnistis citrella*
CLM	Citrus leaf miner
R.H	relative humidity
TM	temperature
SD	standard deviation
r	Correlation coefficient values

CHAPTER – I:

INTRODUCTION & OBJECTIVES

1.1 Introduction

Citrus is infested with many pests that cause severe damage to trees as well as have a significant impact on production. The main pests are: The Mediterranean fruit fly, the red mite, the California louse, aphids, and the citrus leaf miner *Phyllocnistis citrella*. The latter is the most important pest that attacks citrus and other species of the Rutaceae family and some related ornamental plants **(Abbas *et al*., 2013)**.

The native range of *Phyllocnistis citrella* is east and south Asia, including Japan, Thailand, China, India, Malaysia, and Taiwan). It is also present in Australia, Africa, the Mediterranean region, and the Americas **(Urbaneja *et al*. 2000 and Cardwell *et al*., 2008)**. The pest was first found in Libya in 1995 from that time the insect spread rapidly throughout the citrus-growing areas in Libya **(EPPO, 2014).**

The citrus leaf miner is an important factor affecting the production of citrus and causes serious damage to citrus yield because the larvae feed on the leaves and make serpentine mines which affects plant photosynthesis as the larvae consume between 1 and 7 cm^2. Then the edge of the leaves curls upward, followed by chlorosis and later by necrotic spots causing leaf drop **(Knapp *et al*., 1995)**. Additionally, it makes the citrus canker disease worse by giving the bacterium *Xanthomonas axonopodis pv. citri* a point of entrance **(Gottwald *et al*., 1997)**. According to **Jesus *et al*. (2006)**, an increase in the number of CLM mines on the leaf surface causes an increase in the severity of the citrus canker disease.

Several control measurements have been developed in response to the enormous damage that this insect has recently caused. Biological control remains the most popular method for controlling *P. citrella* population. The purpose of any biological control, according to **Amalin *et al*. (2002)**, is to reduce the amount of pest infestations by using natural enemies. Over 40 Hymenoptera species exist, including 25 in Near East countries, attacking citrus leaf miner larvae. No known parasites have been recorded on leaf miner eggs **(Munir., 1996).** Seasonal incidence and population dynamics data are essential in developing pest management methods for this insect. periods of no incidence, beginning of incidence, low incidence, and peak incidence, are important for deciding when to employ management approaches. Many studies have been conducted to study the prevalence of leaf miners in relation to different weather conditions **(Ali and Ali., 2018; Patel *et al*., 2000)** Additionally, *P. citrella* population growth is also significantly influenced by weather, and abiotic variables **(Katole *et al*., 1997)**.

Several studies on the population dynamics of *P. citrella* have been conducted on several citrus cultivars in the Mediterranean Basin region **(Salhi and Doumandji - mitiche.,2009; Ali and Ali., 2018; Gharib *et al*.,2019)**. But rarely in Libya so the present investigation aimed to evaluate the population dynamics of *P. citrella* and its associated parasitoid

1.2 Objectives:

The objectives of this study were to determine:

1- The population fluctuations (natural abundance) of the citrus leafminer, *P. citrella,*

2- Natural abundance and biocontrol efficiency of the parasitoids associated with the citrus leafminer, *P. citrella*, in Surman region.

3- Effect of mean temperature and relative humidity on the population dynamics of *P. citrella* and *S. petiolatus*.

4- Synchronization between *S. petiolatus* and its Insect host.

5- The number of mines/ leaflets on the studied host plants.

CHAPTER – II:

LITERATURE REVIEW

2. literature Review:

2.1. Taxonomic position of Phyllocnistis citrella:

The systematic position of the leafminer is summarized by **Balachowsky (1966)** as follows:

Phylum	Arthropoda
Class	Insecta
Order	Lepidoptera,
Family	Gracillariidae, Synonym, Lithocolletidae,
Genus	*Phyllocnistis*,
Species	*Phyllocnistis citrella* (Stainton), 1856,
Common name	Citrus leaf miner.

2.2. Origin and distribution of *P. citrella*:

The species *P. citrella* is a citrus pest originating from East and South Asia (Japan, Thailand, China, India, Malaysia, Sri Lanka, Bangladesh, Pakistan and Taiwan) **(Urbaneja *et al*. 2000; CABI, 2017)**. It was described for the first time in 1856 in Calcutta, India, by H. T. Stainton. Today, can find it in almost every place where citrus fruits are grown around the world **(Bermudez *et al*., 2004; Vercher *et al*.,2008)**. It rapidly spread to North Africa, the Middle East, and Europe's Mediterranean coast from 1993 to 1995 **(Mafi and Ohbayashi, 2004)**. It was first reported in Australia in the same year **(Beattie and Hardy, 2004)**. It was initially discovered in 1997 in Zimbabwe, Brazil, Colombia, Lebanon, and several Caribbean islands. It was first discovered in western Mexico, Central America, and certain Mexican states in the same year **(CABI, 2021)**. In Hungary, it was discovered in the year 2020 **(Katona *et al*., 2020)**. Right now, *P. citrella* is found in many places where citrus fruits are grown all over the world **(CABI, 2021)**.

In Egypt, Citrus trees in El-Sharkia and Ismailia Governorates were infected by CLM in the early summer of 1994 **(Abdel-Aziz, 1995; Eid, 1998)**. Since then, the pest has expanded rapidly throughout Egypt's citrus farming region.

It was discovered in Algeria in 1994 when it was recorded in the coastal districts of the cities of Mostaganem and Oran **(Berkani, 1995)**. Since then, this Citrus pest has infiltrated all of the country's citrus-growing districts, including Tipaza, Skikda, and Annaba, before spreading to the country's interior, including Blida, Chlef, and Tizi-Ouzou **(Dridi and Berkani, 1996)**.

In 1994, citrus leafminer was found in Tunisia. Since then, it has spread to all the citrus trees in Tunisia and became an economic pest of citrus **(Jerraya *et al*. 1996 and Chermiti *et al*., 1998)**.

In Iraq, the citrus leafminer was discovered between 1992 and 1994. which is considered a serious pest on citrus orchids **(Al-Barak 1994, AI-Jboory et al., 2004).**

In Libya, the Citrus leaf miner was first recorded in 1995 From that time the insect spread rapidly throughout the citrus-growing areas in Libya **(Eppo, 2014 and CABI, 2021)**.

2.3. Host plants of *P. citrella*:

P. citrella is a pest that mostly affects plants in the *Rutaceaea* family and also feeds on plants from different botanical families, including jasmine, mistletoe, willow, and several legumes, where it cannot complete its life cycle. Therefore, its preferred hosts are all members of the Citrus genus including orange, lemon, lime, tangerine, etc. **(Knapp *et al*., 1995; Bermudez *et al*., 2004; Nagamine and Heu, 2002; Godfrey and Grafton-Cardwell, 2002)**.

CLM has been observed to attack various citrus species, including *Citrus aurantium* L., *C. aurantifolia*, *C. maxima*, *C. limon*, *C. medica*, *C. paradise*, *C. sinensis*, *C. reticulata Blanco*, *Aegle marmelos*, *Atalantia*, *Citrofortunella microcarpa*, *Fortunella marginata*, *F. crassifolia*, *Limonia* **(De Prins and De Prins, 2005; 2009)**.

Some fruit trees like grapefruit and mandarin are more sensitive to CLM due to their chemical composition. The number of young flushes on these trees each year can influence the insect's attraction or repellence, affecting the tree's overall vulnerability **(Argove and Rossler, 1996; Rocchini *et al*., 2000; Garcia-Mari et al, 2002)**.

2.4. Damage and symptoms of *P. citrella*:

Females of CLM oviposit on all citrus cultivars' young, tender leaves. On the upper and lower surfaces of the younger leaf, eggs are deposited alongside the midvein. The neonate larva burrows into the leaf tissue and feeds along the midvein, zigzagging its way to the leaf margin, where it pupates **(Uygun *et al*, 2000)**.

Heavy infestation harms the leaves of young shoots, tender stems, and occasionally fruits. This species' larvae prefer the lower surface of the leaves, destroying the epidermis and causing the leaves to curl up and become sclerotic and necrotic. where Larvae create serpentine mines in their hosts' leaves and fruits (rarely), leaving behind frass throughout these mines. This feature helps in separating citrus peel miners and leaf miners **(Jones, 2001).** These mines can make the plant sick and give it a disease called citrus canker **(Riasat et al., 2020)**.

Citrus canker can penetrate the mesophyll of a leaf when citrus leaf miner larvae consume it. The larvae can also disseminate the bacteria within the mine, thereby augmenting the amount of canker inoculums generated in a leaf **(Gottwald *et al*., 2002; Belasque *et al*., 2005)**.

According to **Nawaz *et al*. (2021)**, reducing photosynthesis, premature leaf drop, and reduced development of shoots are consequences of leaf miner damage and eventually cause economic loss by supplying low carbohydrates either to hanging fruits or next-season crops. Economic losses caused by citrus leaf miners in orchards include higher pesticide expenses for treating seedlings and young citrus plants **(De Prins, 2005)**.

2.5. life cycle of *P. citrella*:

2.5.1. The egg stage:

The eggs are flat, oval, transparent in color turning pale yellow after two days of laying, measure 0.2 x 0.3 mm **(Balachowsky, 1966; Guérout., 1974; Quilici *et al*., 1995)**.

Dahmane and Chakali (2020) reported that females tend to lay eggs more frequently on leaves with surfaces smaller than 2 cm^2. Fewer eggs are observed when the surface increases. The female lays around 50 eggs during her lifespan, which can last between 2 and 12 days. It lays eggs singly or in groups of two or three eggs on the upper or lower surface of the leaf primarily near the midrib. Egg laying can also be observed on young branches **(MILL, 2003; Mustafa *et al*., 2014).**

2.5.2. The larval stage:

During its development cycle, *P. citrella* goes through four larval instars the first three of which are only feed, while the fourth stage does not feed **(Guérout., 1974)**.

The first three stages have an oral apparatus typically comprising two dorso-ventrally flattened mandibles flattened blade-like mandibles carrying fine teeth, **(Ayoub, 1960; Balachowsky, 1966; Guérout., 1974; Knapp *et al*., 1995)**.

The third instar CLM larvae transition into a non-feeding pre-pupa stage, while the fourth larval instar folds a leaf piece and spins a silk cocoon for pupation **(Beattie and Hardy, 2004)**.

According to **Boughdad *et al*. (1999)**, the length of larval development varies depending on the stage taken into account. For first, second, and third larval instars, the durations are 2 to 9 days, 2 to 10 days, and 2 to 9 days, respectively.

2.5.3. Pre-Pupal stage:

The leaf miner fourth and last larval stage is a transitional stage between the third stage's active larva and the Pupa, it is yellowish-brown and resembles the third instar larva but it does not feed. According to **Badawy (1967)**, the pupa is a sub cylindrical larva with a lobe containing relict mouthparts and a flattened, nearly quadrangular head capsule, the atrophied oral apparatus in this stage sets it apart from the other three and prevents regular food intake **(Guérout, 1974)**, but which on the other hand uses silk produced from its mouthparts to form a pupal chamber. This chamber is usually located on the leaf margin **(Beattie and Smith, 1993)**.

2.5.4. The Pupal stage:

The pupa is a stage in an insect's life where it transforms into an adult. It starts yellowish-brown with blackish areas, and gets darker as it gets older. Equipped with a sharp frontal spine, undoubtedly intended to perforate the light cocoon formed outside the pupation, this stage takes about 6 to 22 days to finish **(Guérout, 1974; Knapp *et al*., 1995; Kerns *et al*., 2001)**.

2.5.5. The Adult stage:

After 7-10 days of hiding inside a cocoon, the adult emerges and starts moving around. and remains active early in the morning and late in the evening **(Beattie, 1989)**.

The adult moth is silvery-white in color and a size of 4 to 5 mm. A darker line divides the forewings into two sections **(Guérout., 1974)**. The bottom of the forewings is snowy white becoming distally yellow with several small oblique or perpendicular streaks to the rib, in addition to a distal black line forming the ray and a fan of three to four lines of the same color **(Balachowsky, 1966; De Prins and De Prins, 2009)**.

While Long fringe scales that extend from the margins of the hind wings adorn the white body and hind wings. The moth seems much smaller in resting stance, with its folded wings (approximately 2.4 mm). The haustellum lacks basal scales, and the head has very smooth, white scales **(Kawahara *et al*., 2009)**.

Guérout (1974) reported that the apical portion has a marginally distinct color. She has long fringes with three black lines running through them and a distinctive black eyespot at the tip. made up primarily of white, beige, and golden scales with a few black scales. Only the longest, posterior legs have two spurs. not a single character clearly distinguishes between genders. Adults are typically too little to be readily observed, and they are active throughout the day and at night **(De Prins and De Prins, 2009)**.

2.6. Management of *P. citrella*:

2.6.1. Biological control:

Biological control includes, the conservation of natural enemies via orchard management, artificial mass rearing, and release of natural enemies **(Raza *et al*., 2017)** Biological control is a viable and successful long-term solution for managing *P. citrella* **(Kalaitzaki *et al*. 2021)** and classical biological control is often considered a cornerstone of integrated pest management. with the introduction of exotic natural enemies as biological control agents.

According to **Boualem *et al*. (2007)**, three species of parasitoids were recorded in the family Eulophidae: *Pnigalio agraules* (Walker), *Cirrospilus vittatus* Walker, and *Cirrospilus pictus* (*Nees*). In addition to recording three new species: *Citrostichus phyllocnistoides* (Narayanan), *Closterocerus formosus Westwood*, and *Pnigalio pectinicornis* (L.), collected in the Mostaganem region in Algeria.

As part of the campaign against *P. citrella*, *A. citricola*, and *S. petiolatus* were introduced, with the latter being the only species that was able to adapt to local circumstances and was considered to be effective biological control of the leafminer in Tunisia **(Braham *et al*., 2006).**

According to **Khifif *et al*. (2020)**; It has been determined four species of parasitoids in this study in 2017 in Morocco (*S. petiolatus*, *C. phyllocnistoides*, *Cirrospilus pictus Nees* and *Cirrospilus vittatus* Walker (Hymenoptera: Eulophidae) In addition two new species were found in 2018: *Pnigalio* sp. and *Chrysocharis* sp.

2.7. Seasonal abundance of *Phyllocnistis citrella* and its parasitoids:

Jorce *et al*. (1996), studied the seasonal abundance of *P. citrella* and its humenoptrous parasitoids in Florida during the growing seasons of 1993 - 1995 and found that, *P. citrella* density increased from spring through fall and declined during winter 1994 – 1995. They also recorded eight species of Hymenopterous parasitoids attacking *P. citrella* immature stages. The Eulophid *Pnegalio minio* is a primary ectoparasitoid comprised nearly 80% of the parasitoid complex emerged from *P. citrella*, while *Cirrospilus* sp, *Closterocerus* sp, *Zagrammosoma* sp and *Horismenus* sp recorded only 2 – 7% of the parasitoid complex.

Alkhateeb *et al*. (1999) studied the population dynamics of Citrus leafminer *P. citrella* and its parasitoids in Syria, in 1995-1997, weekly random sampling of citrus species revealed maximum infestation levels in July, with leafminer abundance during spring, fall, and summer. August had the highest parasitism rate 73%, with other species like *Ratzeburgiola incomplete*, *Cirrospilus* sp, and *Semielacher petiolatus* also affected.

P. citrella and its parasitoids were examined on eleven citrus species in Ecuador. The highest infestations were found in three locations: Lodana 43.8%, Riochico 45.7%, and La Unión 17.3%. The parasitism rates of *Ageniaspis citricola Logvinovskaya*, *Galeopsomyia* sp, and *Elasmus tischeriae* Howard were 28.4, 2.2, and 0.07%, respectively **(Ernesto *et al*., 2004)**.

Study on *P. citrella* and its parasitoids in Iraq from 2004 to 2005 revealed that insect larvae abundance peaks during cold months, while pupa density peaks in April. The insects produce 11-12 generations annually, with two parasitoids population peaks: October and July. The dominant parasitoids were *Ratzeburgiola incompleta Boucek*, *Cirrospillus* sp., *Neochrysocharis formosa*, *Pnigalio* sp., and *Baryscapus* sp., while *Tetrastichus* sp. and *Pediobius* sp. were found in small numbers **(Aljboory *et al*, 2004)**.

Shebli and Kerra (2006), surveyed *P. citrella* on different varieties of citrus trees in Libya in 2005. They found the infestation rate in Ain Zara and Zawia reached 99.0% and 96.7%, respectively, on the new shoots in the fall season. The highest population of the insect was found on lemon trees, while the lowest population was found on the Hasna tree variety.

The study conducted in Tafí Viejo, Tucuman province, found that citrus leafminer populations increased in spring and summer, decreased in fall, and disappeared in winter. Five parasitoid species, including four native and one exotic, preyed on the juvenile stages of citrus leafminers, with the highest rates observed in fall **(Diez *et al*, 2006)**.

Salhi and Doumandji -mitiche. (2009), conducted a study to determine the population fluctuations of *P. citrella* and its parasitoids in Algeria and found the citrus leaf miner develops three generations, mainly in summer and autumn flushes. three Hymenoptera Eulophidae associated with *P. citrella* were recorded: *Pnigalio* sp. and *Cirrospilus pictus Nees*, native ectoparasitoids: and *Ageniaspis citricola Logvinovskaya*, an exotic endoparasitoid.

Seasonal population dynamics of *P. citrella* (Stainton) in Sudan were studied on four citrus species: grapefruit, orange, local lime, and mandarin. Results showed that the peak population in december to february coincided with the availability of new vegetable flush leaves, with grapefruit being the most susceptible to leaf miners, followed by orange **(Ali and Ali, 2018)**.

Gharib *et al*. (2019) studied the population dynamics of *P. citrella* and its associated parasitoids on mandarin trees during two successful years 2013-14 & 2014-15 in Sharkia Governorate, Egypt. and found that the citrus leafminer larval population had 6 peaks of abundance on mandarin trees in each studied year. The higher population in late May, late August and the middle of September and recorded five peaks of parasitism, the highest parasitism rate was 42.5% at the beginning of august and 19.2% in the middle of september two parasitoids were identified including *S. petiolatus* and *Cirrospilus ingenuus* (Gahan).

Sharma and Khokhar (2019) study on *P. citrella* (Stainton) seasonal population dynamics from january 2016 to august 2018 found that citrus leaf miners had two peaks of infestation in Punjab. The population was higher in summer than autumn, starting in february and reaching a peak in May and september before declining. Climatic conditions were found to be important factors in determining the intensity of *P. citrella* incidence.

2.8. The ectoparasitoid *semielacher petiolatus*: -

Semielacher petiolatus (Girault) (Hymenoptera Eulophidae) is a solitary ectoparasitoid, indigenous to Australia, it is the only species known as a *P. citrella* parasitoid that has a funiculus with two articles in both sexes and a very distinct petiole **(Schauff *et al*., 1998)**.

S. petiolatus parasitoid has been introduced into many Mediterranean nations, including Greece, for the biological control of citrus leafminer, *S. petiolatus* was present from mid-june to early december, while its population was very low in the fall dispersing very rapidly over large contributing to a reduction in the citrus leafminer population **(Kalaitzaki *et al*., 2011)**.

Harbi *et al*. (2018) estimate the parasitism rate caused by endemic and imported parasitoid species associated with *P. citrella* in three citrus-growing regions in Tunisia. The results indicated that the activity of *S. petiolatus* started in July with a maximum parasitism rate of 3.33% recorded in September while the total parasitism rate ensured by *S.*

petiolatus was about 57% the impact of *S. petiolatus* on the larval populations of *P. citrella* was low.

According to **Elekçioğlu and Uygun (2006)**, there were no *S. petiolatus* specimens in 1996, even at release locations. However, even far from the releasing location, 100 *P. citrella* individuals were discovered in samples that were taken in 1997 from different locations in the eastern Mediterranean region. this finding suggested that the parasitoid established itself in Turkey's eastern Mediterranean citrus-growing region. However, this parasitoid was only seen in extremely small amounts in the years that followed.

2.9. Effect of mean temperature and relative humidity on the population dynamics of *P. citrella* and *S. petiolatus*:

Kumbhar *et al*, (2021) studied the relationship between abiotic factors with population dynamics of citrus leaf miner, *P. citrella* on acid lime and found that the larval population and percent leaf infestation were positively correlated with minimum temperature, morning relative humidity, and evening relative humidity.

Prabhudev *et al*., (2021) reported that the correlation of weather parameters with citrus leaf miner incidence indicated that rainfall, minimum and maximum temperature, and minimum relative humidity had a non-significant effect. In contrast, maximum relative humidity had a positive and significant impact.

Chhetry, (2012) concluded that correlation analysis indicated that the percentage infestation was significantly correlated with maximum temperature and minimum temperature. Average rainfall showed a positive correlation with the population, but not average relative humidity, it also showed results from **Arshad *et al*. (2021)** The maximum and minimum temperatures were found to be significantly and positively correlated with *P. citrella* infestation ($P < 0.05$), but relative humidity showed a negative correlation with *P. citrella* infestation at $P>0.05$, during both years. so, temperature stands out as the most important determinant constraining *P. citrella*.

Anjitha *et al*, (2014) studied the effect of weather parameters on the incidence of citrus leaf miner, and its natural enemies in three commercially grown citrus cultivars and found that maximum temperature have a significant negative correlation towards the CLM infestation as well as parasitization rate on the three cultivars, while relative humidity was found to have a significant positive role in favoring the incidence of CLM.

Kalaitzaki *et al*. (2018) studied the effect of temperature on the development and survival of the parasitoid *S. petiolatus* (Girault) on its host on two important Greek citrus cultivars and found that parasitoid development was favored at temperatures ranging from 20-30°C but not at the highest temperature tested (32.5°C). The failure of *S. petiolatus* to complete its development at a constant temperature of 32.5 °C, where the shortest developmental period was recorded at 30 °C and the longest at 15 °C. No adults emerged at 32.5 °C.

Patel *et al*. (2000) investigated the influence of temperature on the preimaginal development of *P. citrella* on kagzilime and discovered that temperatures above 10°C had a substantial effect on the duration of egg, larval, pupal, and life span of *P. citrella*. In comparison to lower 10°C and 15°C or higher 30°C and 35°C temperatures, a much higher percentage of larvae could pupate at 20°C and 25°C temperatures. Male and female moths lived longer in lower temperatures than in higher temperatures. Female moths that emerged from larvae maintained at 25°C had the highest fecundity of 52 eggs per female, but at 35°C, 17 eggs per female were found *P. citrella* grew in ideal conditions at 25°C.

CHAPTER – III:

MATERIALS AND METHODS

3. Materials and methods:

3.1. study Area:

To estimate the population fluctuation of citrus leafminer *P. citrella* and the incidence of its parasitoids, weekly samplings were collected during the period from May 2018 to March 2019 from a citrus orchard drip-irrigated grove in the Surman region, (Location: Latitude 32.7562 Longitude 12.5693) Planted with the following trees: different species of citrus, date palms, and grapes, the orchard covers 1 hectare, that orchard did not receive any insecticidal treatments during the study.

3.2. Host plants:

For this investigation, four citrus varieties were targeted: Lemon (*Citrus limon*), Hasna or blood orange (*Citrus sinensis*), Abu sora, Washington navel (*Citrus sinensis*(*osbeck*)) and Tarocco orange (*Citrus sinensis*).

3.3. Sampling:

In each sample, five trees were randomly selected from each citrus species. The canopy of each tree was divided into two sides (north and south) two layers (one and two meters above the ground) and one flush, where 20 young leaves were collected from each tree. Therefore, 100 young leaves per citrus species were collected in each sample weekly and placed into plastic bags until examination, where 12,100 leaves were examined in this study from all citrus species were collected.

3.4. Examination:

Leaves were examined under a binocular stereo-microscope for the presence of mines (either occupied or abandoned), larvae (first to fourth instar based on their morphology), and pupae of the citrus leafminer, live and dead, as well as parasitoid Immature stages (eggs, larvae, pupa). The results were recorded in weekly tables for each month, Leaves containing parasitized individuals of *P. citrella* were placed in Petri dishes with water-soaked cotton, until the adult emergence of the parasitoids. Adult parasitoids were collected in plastic vials and kept for identification, Parasitoid *S. petiolatus* were identified by sent to Dr Alansary. R. Elkhouly who confirmed the identification.

3.5. metrological studies:

Daily records of mean temperature along with relative humidity obtained from the meteorological station of Tripoli to represent the climatic condition effect, mean values of temperature, and relative humidity were calculated, according to the following model.

$$Tm = \frac{Tmax + Tmin}{2}$$

3.6. statistical analysis:

The arithmetic averages, standard deviation, coefficient correlation values, and regressions were estimated using Microsoft Excel software 2016, Statistical analysis of the differences between the monthly averages was conducted using the T-Test at the threshold of 0.05.

CHAPTER – IV:

RESULTS

4. Results:

In total, 12,100 leaves were inspected between May 2018 and March 2019 to determine the presence or absence of *P. citrella* immature stages and its parasitoids in the citrus orchard in the Surman region were 4000 leaves of lemon, 3300 leaves of abu sora, 2400 leaves of tarocco, and 1900 leaves of hasna examined.

4.1. Seasonal abundance of *Phyllocnistis citrella*

4.1.1: - On Lemon *Citrus Limon (L)*Osbeck

4.1.1.1. *P. citrella* larval stage:

As presented in fig (1), *P. citrella* larvae recorded low numbers at the beginning of the season in early June then the population increased recording four peaks of abundance (185, 206, 212, and 288 individuals /100infested leaves) occurred on the 21st of July the 18th of August,17th of November and 1st of December respectively, while dead larvae recorded five peaks of abundance (82, 64, 97, 119, and 92 individuals/100 infested leaves) occurred on the 8th of September, the 27th of October, the 24th of November, the 1st of December, and the 8th of December, the total of living larvae and dead of *P. citrella* in Lemon recorded six peaks of abundance (203, 255, 236, 230, 312, and 407 individuals /100 infested leaves) occurred on the 21st of July, the 18th of August, the 1st of September, the 27th of October, the 24th of November, and the 1st of December.

4.1.1.2. *P. citrella* Pre Pupal stage:

As shown in Fig (2) Living Pre pupa of *P. citrella* on Limon recorded four peaks of abundance (49, 19, 12, and 14 individuals /100 infested leaves) occurred on the 7th of July, the 25th of August, the 27th of October, and the 1st of December, while the dead Pre pupa recorded two peaks of abundance (16, and 17 individuals /100 infested leaves) occurred on the 27th of October, and the 9th of March. On the other hand, the total of Pre pupa of *P. citrella* recorded four peaks of abundance (49, 21, 28, and 26) that occurred on the 7th of July, the 25th of August, the 27th of October, and the 9th of March.

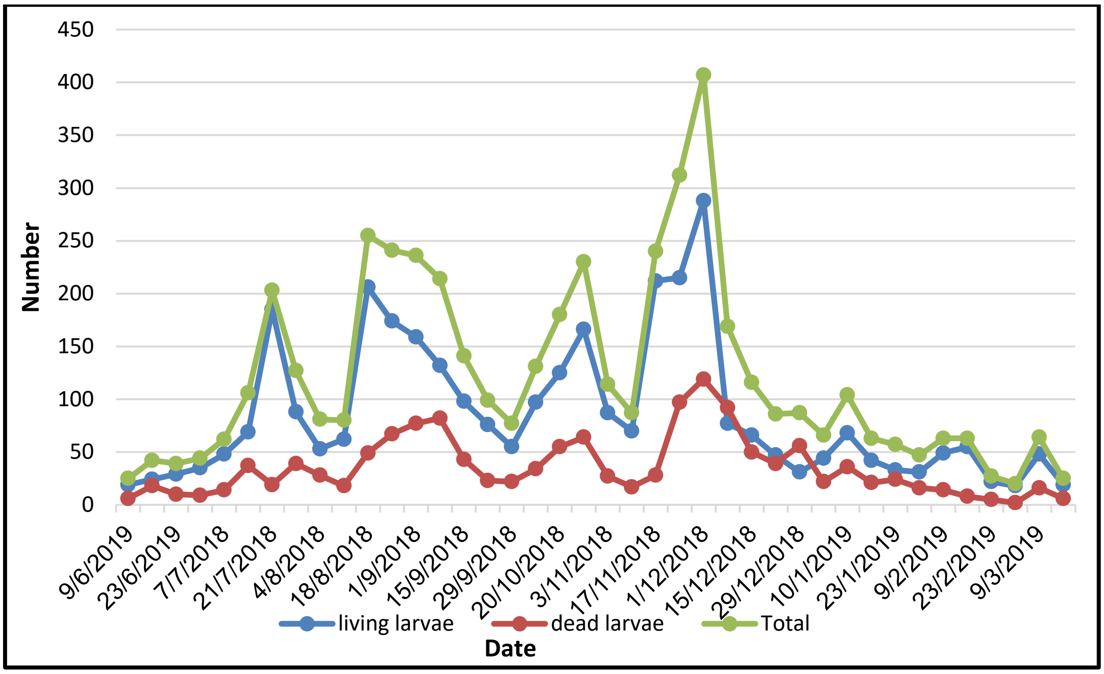

Fig (1) Population abundance of living larvae, dead larvae and the total of *P. citrella* on Lemon during season 2018-2019.

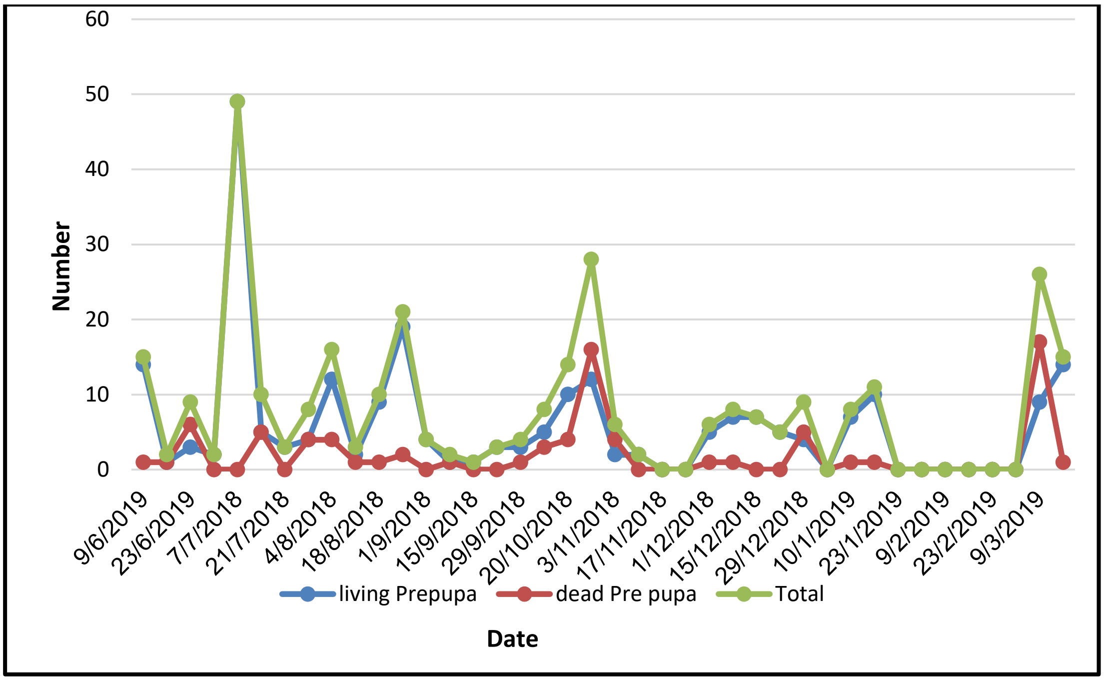

Fig (2) Population abundance of living pre pupa, dead pre pupa and the total of *P. citrella* on lemon during season 2018-2019.

4.1.1.3. *P. citrella* Pupa stage:

As presented in Fig (3), *P. citrella* pupa recorded five peaks of abundance (23, 29, 66, 47, and 24 Individuals / 100 infested leaves) occurring on the 16th of June, 30th of June, 10th of January, 17th of January, and 8th of March, while the dead pupa recorded two peaks of abundance (14 and 14 individuals/100 infested leaves) occurred on the 9th of June, and the 16th of March. As for as The total number of pupa of *P. citrella* recorded five peaks of abundance (29, 22, 71, 51, and 31 individuals /100 infested leaves) on the 30th of June, the 27th of October, the 10th of January, the 17th of January, and the 9th of March.

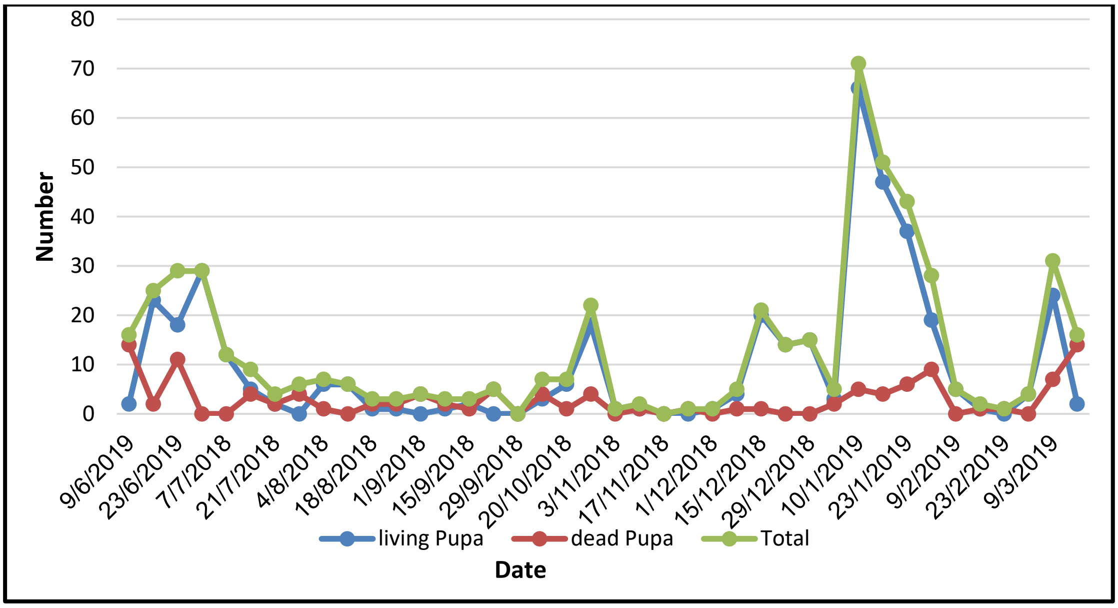

Fig (3) Population abundance of living pupa, dead pupa and the total of *P. citrella* on lemon during season 2018-2019

As shown in Table (1) *P. citrella* stages on lemon showed their highest monthly average numbers in November (188.25 ± 106.08 individuals / 100 infested leaflets) for larvae and 17.5±21.21 individuals / 100 infested leaflets in July for pre pupa and 39.6±24.80 individuals / 100 infested leaflets in January for pupa stage. On the other hand, *P. citrella* stages on lemon showed their lowest monthly average numbers occurred in March 36.33±24.091ndividuals / 100 infested leaflets for larvae and 0 individuals / 100 infested leaflets in February for pre pupa and 1± 0.82 individuals / 100 infested leaflets in November for pupa stage.

Table (1) Total average numbers ± SD of CLM stages and infested leaflets during the period of the study on *Citrus Limon* (L) Osbeck and T.test values

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T test
June	37.5 ± 8.58	7.0 ± 6.27	24.75 ± 6.13	2.45
July	124.75 ± 95.37	17.5 ± 21.21	7.75 ± 3.5	1.29
August	164.25 ± 96.88	12.5 ± 7.77	4.5 ± 2.06	1.98
September	153.4 ± 69.72	2.8 ± 1.30	3.0 ± 1.87	1.67
October	180.33 ± 49.50	16.67 ± 10.26	12.0 ± 8.66	2.55

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T test
November	188.25 ± 106.08	2.0 ± 2.83	1.0 ± 0.82	2.34
December	173.0 ± 135.08	7.0 ± 1.58	11.2 ± 8.08	2.56
January	67.4 ± 21.71	3.8 ± 5.31	39.6 ± 24.80	2.23
February	51.0 ± 20.78	-	2.67 ± 2.08	2.45
march	36.33 ± 24.09	13.67 ± 13.05	17.0 ± 13.53	2.27
Mean ± S.d	117.62 ± 59.18	8.293± 6.96	12.37167 ± 7.15	2.23

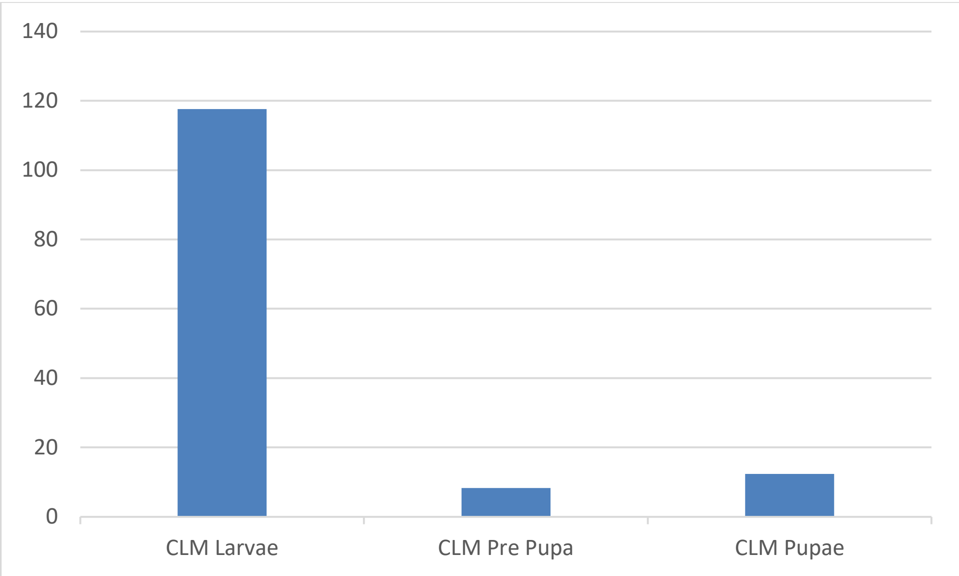

Fig (4) Total average number of CLM stages and infested leaflets during the period of the study on Lemon

4.1.2: - On Abu sora, Washington navel (*Citrus sinensis osbeck*)

4.1.2.1. *P. citrella* larvae stage:

As presented in Fig (5), *P. citrella* larvae on Abu sora Washington navel recorded five peaks of abundance (184, 313, 279, 255 and 189 individuals/100 infested leaves) occurred on 14^{th} of July, 25^{th} of August, 8^{th} of September, 22^{th} of September and 6^{th} of October respectively, while dead larvae recorded three peaks of abundance (113, 102, and 104 individuals /100 infested leaves) on the 1^{st} of September, the 22^{th} of

September, and 6th of October. On the other hand, the total number of *P. citrella* larvae recorded six peaks of abundance (232, 224, 369, 365, 357, and 293 individuals /100 infested leaves) on the 14th of July, the 11th of August, the 25th of August, the 8th of September, the 22th of September and the 6th of October.

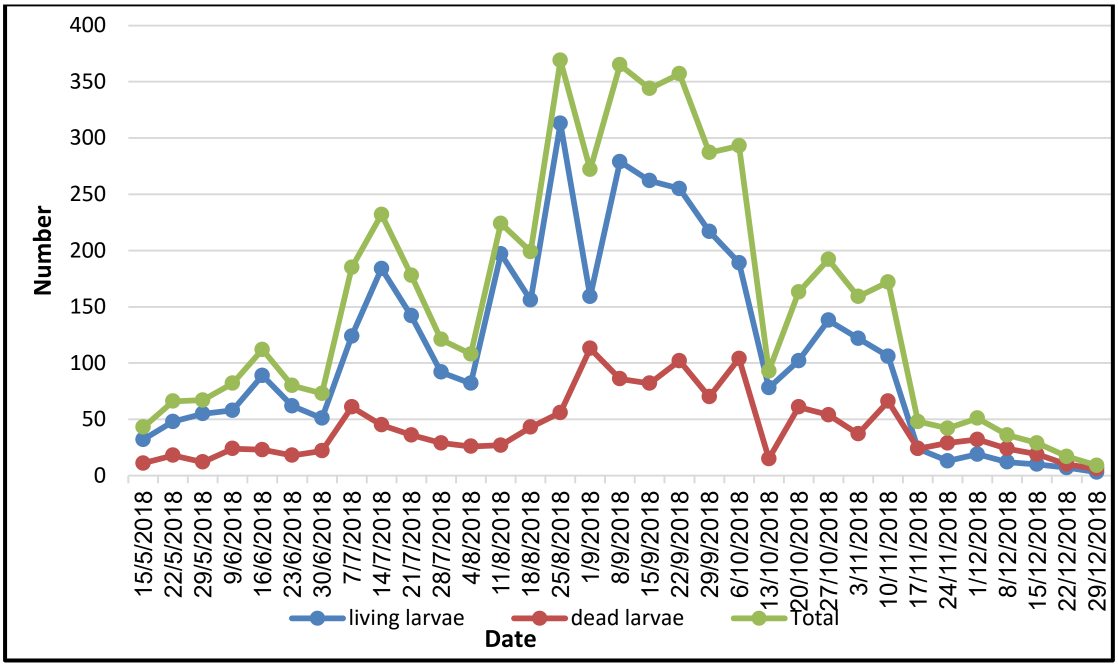

Fig (5) Population abundance of living larvae, dead larvae, and the total of *P. citrella* on Abu sora Washington navel during season 2018-2019.

4.1.2.2. *P. citrella* Pre pupa stage:

As presented in Fig (6), The population of living *P. citrella* pre pupa recorded three peaks of abundance (23, 11, and 11 individual/ 100 infested leaves) on the 7th of July, 14th of July, and 20th of October, while dead pre pupa recorded three peaks of abundance (12, 6, and 33 individuals /100 infested leaves) on the 22nd of May, the 4th of August, and the 27th of October. As for, the total number of *P. citrella* pre pupa recorded three peaks of abundance (15, 28, and 33 individuals /100 infested leaves) occurred on the 22nd of May, the 7th of July, and the 27th of October.

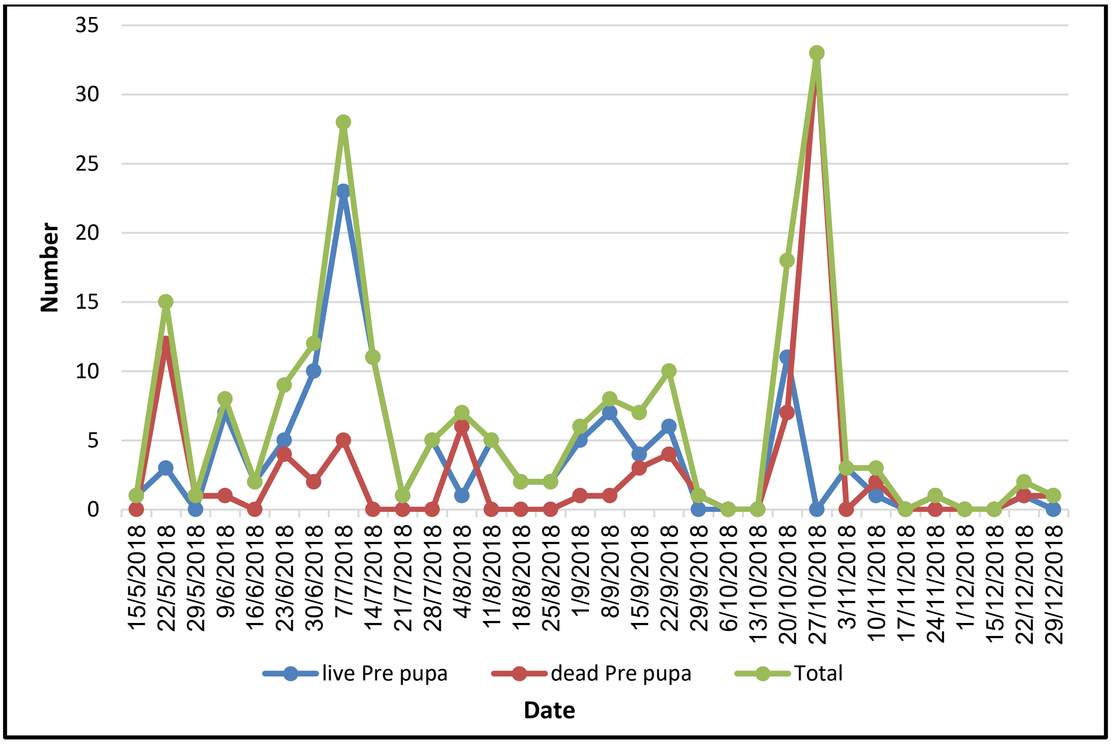

Fig (6) Population abundance of Pre pupa, dead Pre pupa and the total of *P. citrella* on Abu sora Washington navel during season 2018-2019

4.1.2.3. *P. citrella* Pupa stage:

As presented in Fig (7), *P. citrella* pupa recorded the highest number at the beginning of the season where five peaks of abundance 20, 34, 38, 62, and 22 individual/ 100 infested leaves on the 22nd of May, 9th of June, 16th of June, 30th of June and 7th of July respectively, while the dead pupa recorded one peak of abundance (18 individuals /100 infested leaves) on the 27th of October. As for, the total of *P. citrella* pupa recorded five peaks of abundance (25, 37, 39, 62, and 28 individuals /100 infested leaves) occurred on the 22nd of May, the 8th of May, the 16th of May, the 30th of May, and the 27th of October.

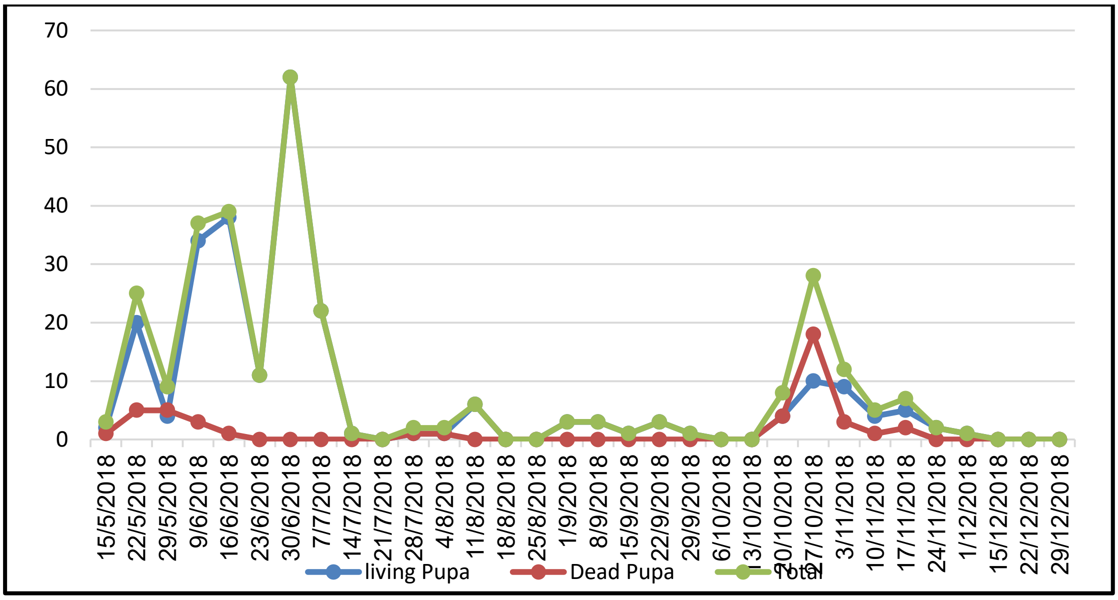

Fig (7): Population abundance of living pupae, dead pupae and the total of *P. citrella* on Abu sora Washington navel during season 2018-2019.

As shown in Table (2) *P. citrella* stages on Washington navel (*Citrus sinensis Osbeck*) showed their highest monthly average numbers in September 325±42.54 individuals / 100 infested leaflets for larvae and 12.75±15.95 individuals / 100 infested leaflets in October for Pre pupa and 37.25±20.86 individuals / 100 infested leaflets in June for pupae stage. On the other hand, as shown in Table (2) *P. citrella* stages on Washington navel showed their lowest monthly average numbers in December 28.4±16.40 individuals / 100 infested leaflets for larvae and 0.8±0.84 individuals) / 100 infested leaflets in December for pre pupa and 0.2±0.45 individuals / 100 infested leaflets in December for pupae stage.

Table (2): Total average numbers ± SD of CLM stages and infested leaflets during the period of the study on Washington navel (*Citrus sinensis osbeck*) and T.test values

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T Test
May	58.67 ± 13.58	5.67 ± 8.08	12.33 ± 11.37	2.55
June	86.75 ± 17.27	7.75 ± 4.19	37.25 ± 20.86	2.23
July	179.0 ± 45.50	11.25 ± 11.90	6.25 ± 10.53	2.45
August	225.0 ± 108.17	4.0 ± 2.45	2.0 ± 2.83	2.11

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T Test
September	325.0 ± 42.54	4.6 ± 3.36	2.2 ± 1.10	2.93
October	185.25 ± 82.90	12.75 ± 15.95	9.0 ± 13.22	2.19
November	105.25 ± 69.82	1.75 ± 1.5	6.5 ± 4.20	2.23
December	28.4 ± 16.40	0.8 ± 0.84	0.2 ± 0.45	1.89
Mean ± S.d	149.17 ± 49.53	6.30 ± 6.03	9.47 ± 8.07	2.11

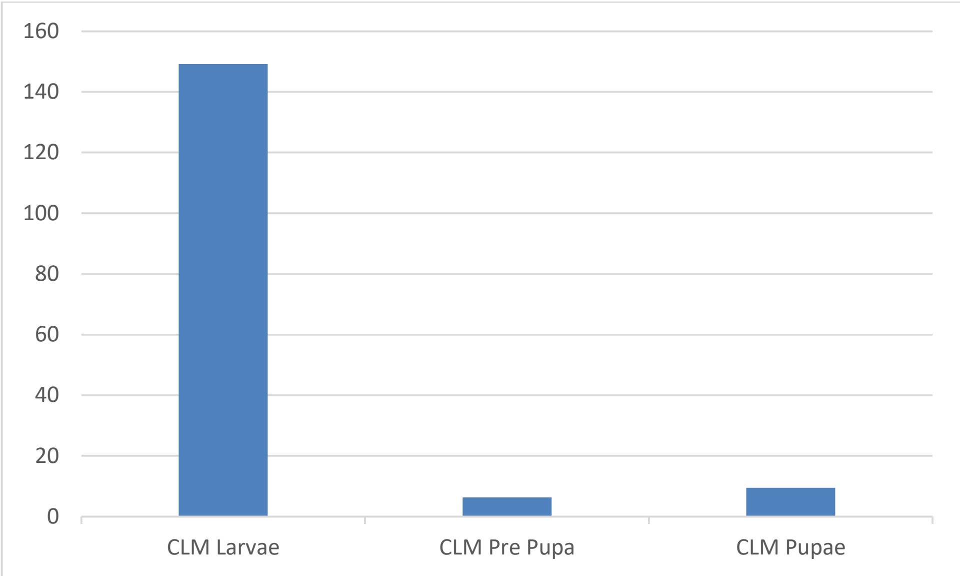

Fig (8) Total average number of CLM stages and infested leaflets during the period of the study on Abusora

4.1.3: - On Tarocco orange (*Citrus sinensis*).

4.1.3.1. *P. citrella* larvae stage:

As presented in Fig (9), *P. citrella* larvae recorded high seasonal abundance during most of the study period and recorded five peaks of abundance (87, 139, 259, 212, and 107 individuals/100infested leaves) occurred on the 30th of June, the 21st of July, the 1st of September, the 15th of September, and the 10th of November, while the dead larvae recorded four peaks of abundance (66, 98, 87, and 145 individuals /100 infested leaves) on the 21th of July, the 25th of August, the 8th of September, and the 15th of September. On the other hand, the total number of *P. citrella* larvae recorded seven peaks of abundance (126, 205, 244, 316, 326, 357, and 154 individuals /100 infested leaves) occurred on the 30th of June, the 21st of July, the 18th of August, the 25th of August, the 1st of September, the 15th of September, and the 10th of November respectively.

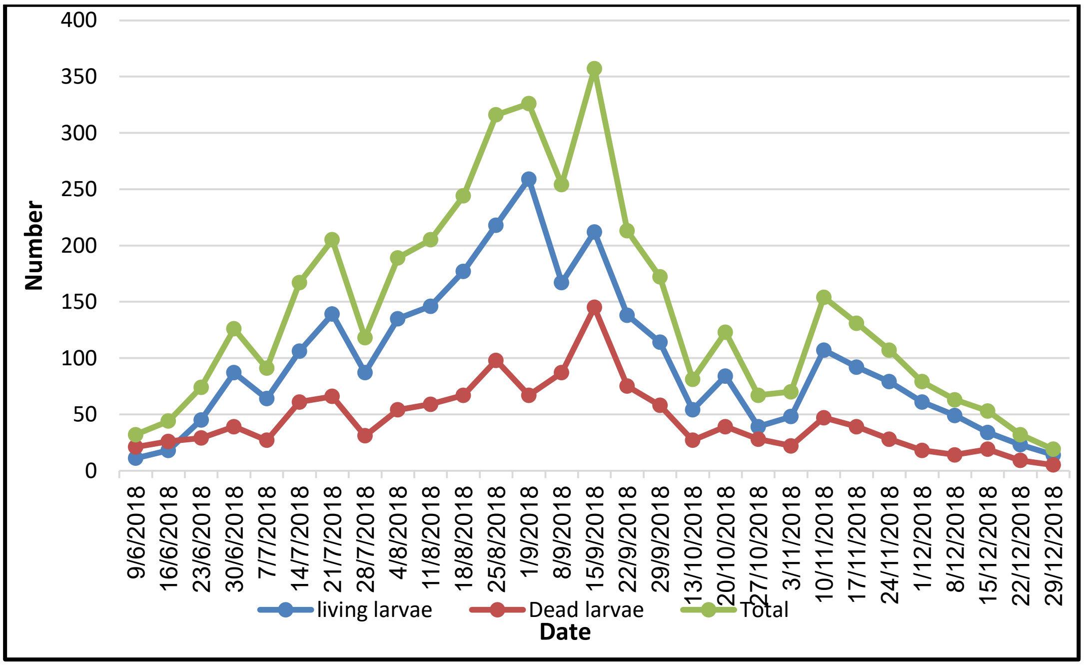

Fig (9) Population abundance of living larvae, dead larvae and the total of *P. citrella* on Tarocco orange during season 2018-2019.

4.1.3.2. *P. citrella* Pre-pupa stage:

The pre-pupa stage population as presented in Fig (10), recorded three peaks (10, 19, and 16 individuals /100 infested leaves) occurring on the 30th of June, the 7th of July, and the 20th of October, while the dead pre pupa recorded one peak (9 individual /100 infested leaves) on the 20th of October. On the other hand, the total number of *P. citrella* pre pupa recorded two peaks of abundance (19, and 25 individuals /100 infested leaves) occurred on the 7th of July, and the 20th of October.

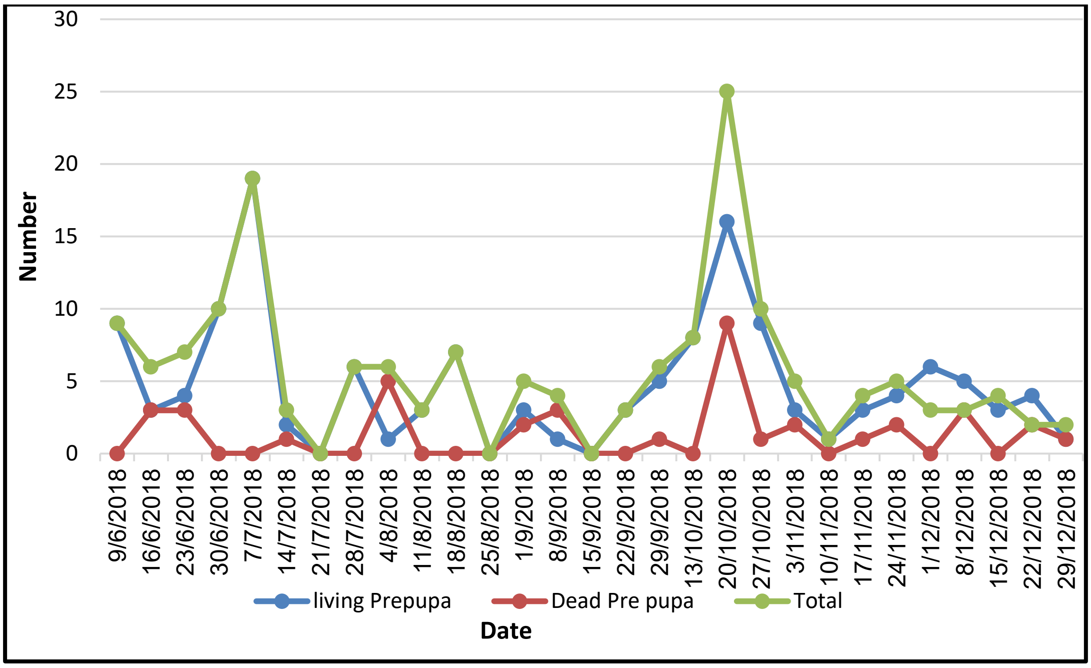

Fig (10) Population abundance of living pre pupa, dead, and the total of *P. citrella* on Tarocco orange during season 2018-2019.

4.1.3.3. *P. citrella* Pupae stage:

As presented in Fig (11), the *P. citrella* pupae living population, however, recorded four peaks of abundance (47, 33, 16, and 25 individual/100 infested leaves) occurring on the 9th of June, the 30th of June, the 20th of October, and the 27th of October respectively, while the dead pupa didn't any peaks of abundance. However, the total of pupa recorded four peaks of abundance (52, 47, 30, and 30 individuals /100 infested leaves) occurred on the 9th of June, the 16th of June, the 30th of June, and the 27th of October.

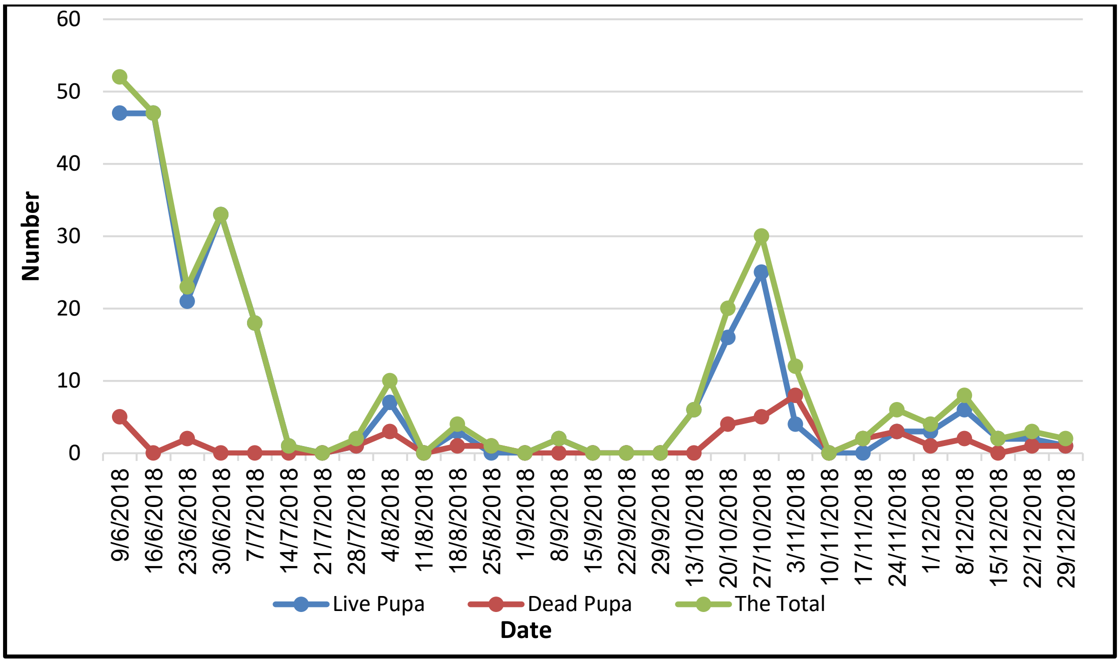

Fig (11) Population abundance of living pupa, dead pupa and the total of *P. citrella* on Tarocco orange during season 2018-2019.

As shown in Table (3) *P. citrella* stages on Tarocco orange (*Citrus sinensis*) showed their highest monthly average numbers in September (264.4±76.91 individuals /100 infested leaflets) for larvae and (14.33±9.29 individuals / 100 infested leaflets) in October for pre pupae and 38.75±13.23 individuals / 100 infested leaflets in June for pupae stage. On the other hand, *P. citrella* stages showed their lowest monthly average numbers in December 49.2±23.98 individuals /100 infested leaflets for larvae and 2.8±0.84 individuals /100 infested leaflets in December for pre pupae and 0.4±0.89 individuals/100 infested leaflets in September for pupal stage.

Table (3) Total average numbers ± SD of CLM stages and infested leaflets during the period of the study on Tarocco orange and T.test values.

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T Test
June	96.0 ± 41.91	8.0 ± 1.83	38.75 ± 13.23	2.52
July	145.25 ± 50.76	7.0 ± 8.37	5.25 ± 8.54	2.41
August	238.5 ± 56.60	4.0 ± 3.16	3.75±4.5	2.33
September	264.4 ± 76.91	3.6 ± 2.30	0.4 ± 0.89	2.11
October	90.33 ± 29.14	14.33 ± 9.29	18.67 ± 12.06	1.69

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupae Mean ± S.d	T Test
November	115.5 ± 35.90	3.75 ± 1.89	5.0 ± 5.29	2.45
December	49.2 ± 23.98	2.8 ± 0.84	3.8 ± 2.49	1.95
Mean ± S.d	138.88 ± 45.03	6.213 ± 3.95	10.80 ± 6.71	2.28

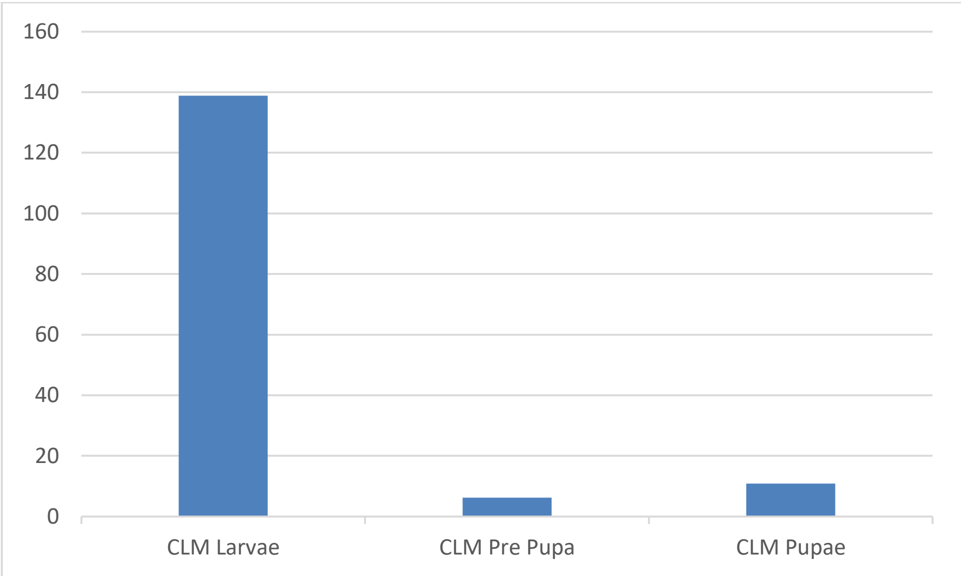

Fig (12) Total average number of CLM stages and infested leaflets during the period of the study on tarocco

4.1.4: - On Hasna (*Citrus sinensis*)

4.1.4.1. *P. citrella* larvae stage:

As shown in Fig (13) Living *P. citrella* larvae recorded four peaks of abundance (137, 156, 194, and 180 individuals/100 infested leaves) occurring on the 14th of July, the 11th of August, the 25th of August, and the 1st of September. While the dead larvae recorded three peaks of abundance (63, 52, and 67 individuals/100 infested leaves), occurring on the 14th of July, the 11th of August, and the 1st of September, respectively. As for the total number of *P. citrella* larvae recorded, there were four peaks of abundance (200, 208, 233, and 247 individuals/100 infested leaves), occurring on the 14th of July, the 11th of August, the 25th of August, and the 1st of September.

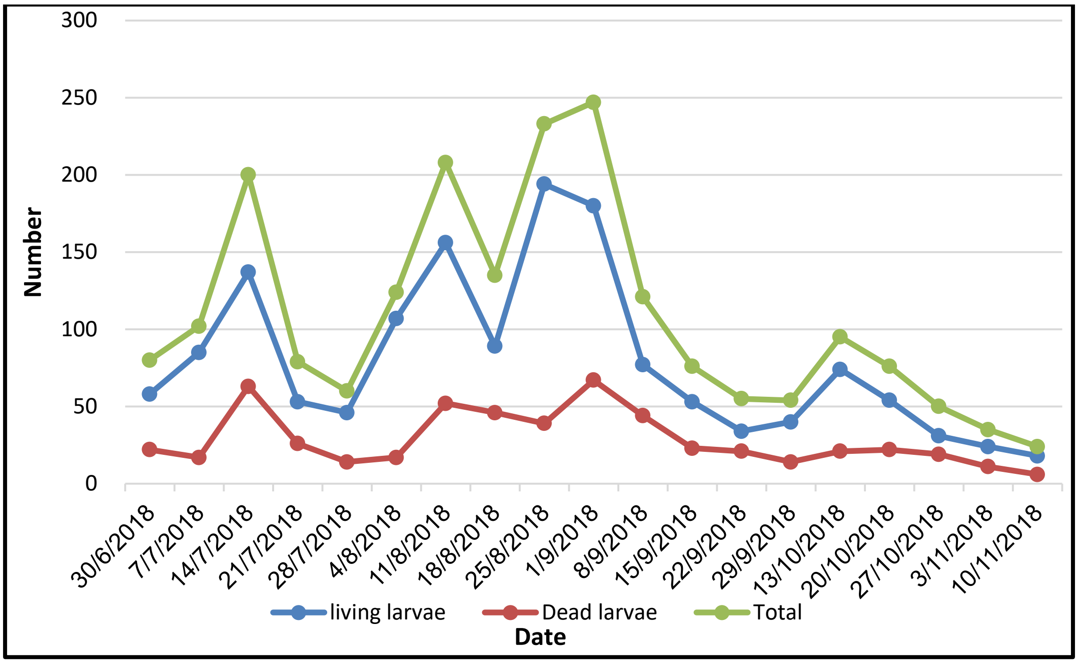

Fig (13) Population abundance of living larvae, dead larvae, and the total of *P. citrella* on Hasna (*Citrus sinensis*) during season 2018-2019.

4.1.4.2. *P. citrella* Pre-pupa stage:

As presented in Fig (14), living pre-pupa of *P. citrella* recorded three peaks of abundance (20, 11, and 12 individual /100 infested leaves) occurring on the 7th of July, the 11th of September, and the 20th of October, while dead pre-pupa recorded one peak of abundance (3 individuals/100 infested leaves) occurred on the 8th of September, As for the total number of *P. citrella* pre-pupa recorded three peaks of abundance (21, 11, and 13 individuals /100 infested leaves) occurring on the 7th of July, the 11th of August, the 20th of October.

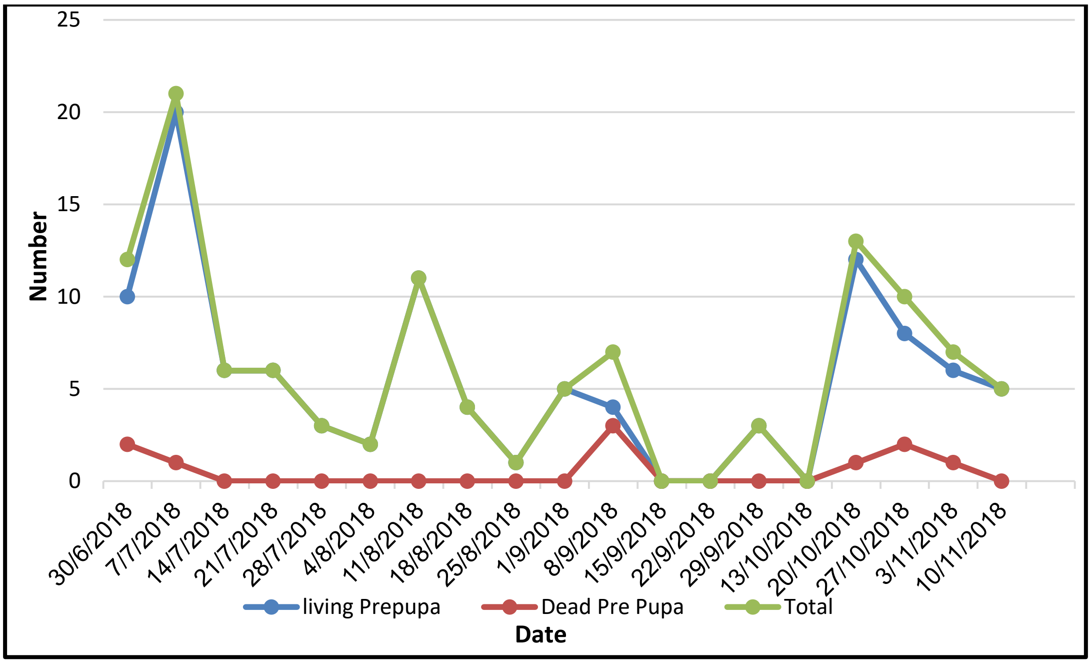

Fig (14) Population abundance of living Pre-pupa, and dead Pre-pupa and the total of *P. citrella* on Hasna (*Citrus sinensis*) during season 2018-2019.

4.1.4.3. *P. citrella* Pupal stage:

As presented in Fig (15) living pupae of *P. citrella* recorded three peaks of abundance (12, 19, and 9 individuals /100 infested leaves) occurring on the 30th of June, the 7th of July, and the 20th of October, while the dead pupa didn't record any peaks of abundance. On the other hand, for the total number of them recorded three peaks of abundance (12, 19, and 11 individuals /100 infested leaves) occurred on the 30th of June, the 7th of July, and the 20th of October respectively.

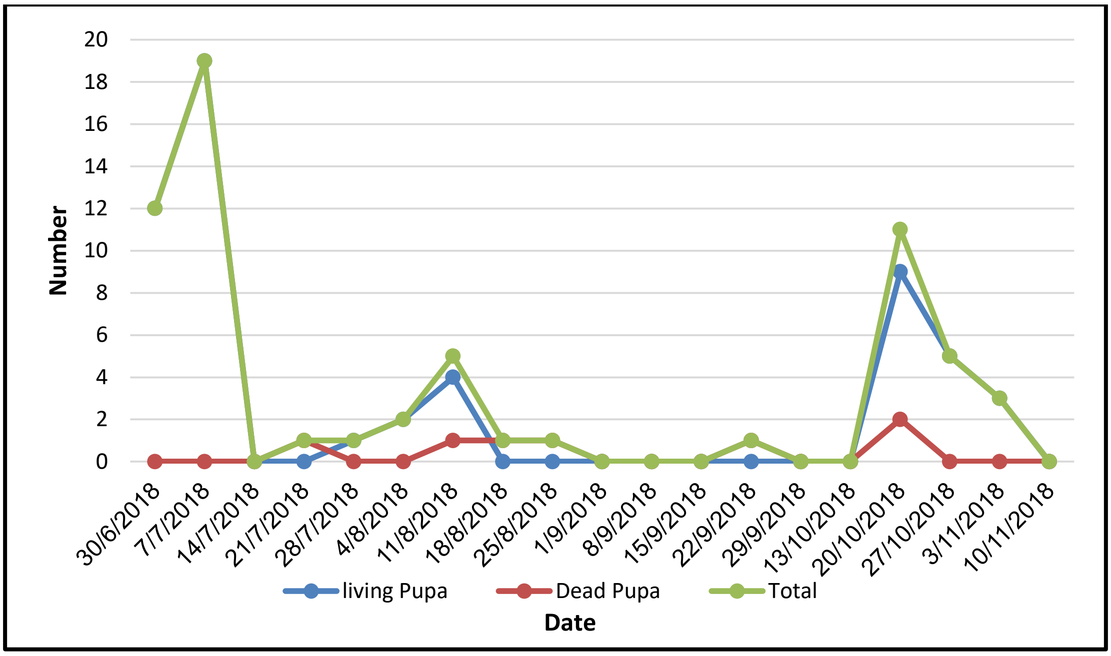

Fig (15) Population abundance of living pupa, dead pupa, and the total pupa of *P. citrella* on Hasna (*Citrus sinensis*) during season 2018-2019.

As shown in Table (4) *P. citrella* stages on Hasna (*Citrus sinensis*) showed their highest monthly average numbers in August 175±53.71 individuals / 100 infested leaflets for larvae and 9±8.12 individuals / 100 infested leaflets in July for pre pupa and 8.75±3.59 individuals / 100 infested leaflets in June for pupa stage. On the other hand, shown in Table (3) *P. citrella* stages on Hasna showed their lowest monthly average numbers in November 29.5±7.78 individuals /100 infested leaflets for larvae and 3±3.08 individuals /100 infested leaflets in September for pre pupa and 0.2±0.45 individuals / 100 infested leaflets in September for pupae stage.

Table (4) Total average numbers ± SD of CLM stages and Infested leaflets during the period of the study on Hasna (*Citrus sinensis*) and T.test values.

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupa Mean ± S.d	T Test
June	127.5 ± 41.56	7.0 ± 5.23	8.75 ± 3.59	1.75
July	110.25 ± 62.25	9.0 ± 8.12	5.25 ± 9.18	2.12
August	175.0 ± 53.71	4.5 ± 4.51	2.25 ± 1.89	2.45
September	110.6 ± 80.94	3.0 ± 3.08	2.0 ± 0.45	1.67

Months	CLM Larvae Mean ± S.d	CLM Pre Pupa Mean ± S.d	CLM Pupa Mean ± S.d	T Test
October	73.67 ± 22.59	7.67 ± 6.81	5.33 ± 5.51	2.31
November	29.5 ± 7.78	6.0 ± 1.41	1.5 ± 2.12	2.78
Mean ± S.d	104.42 ± 44.80	6.19 ± 4.86	3.88 ± 3.79	2.23

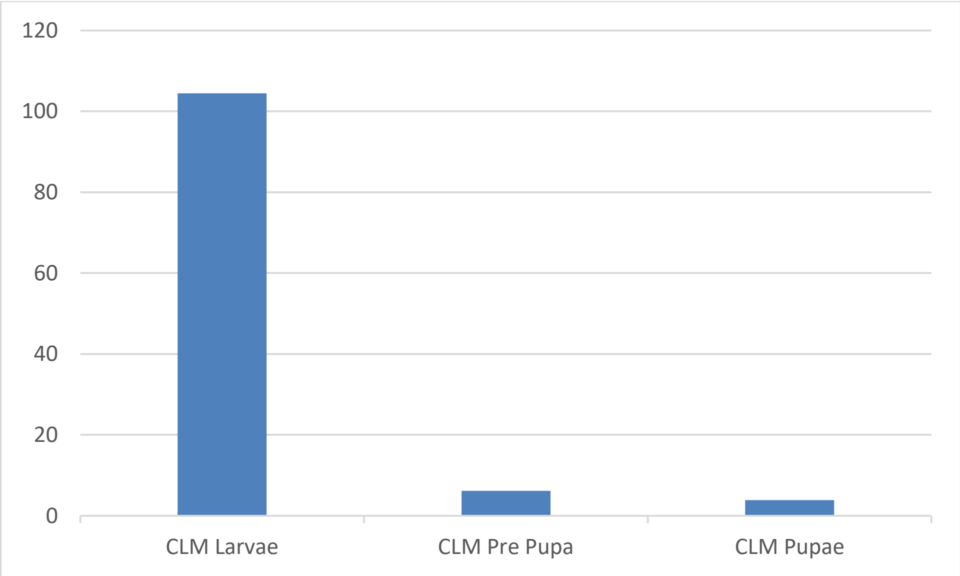

Fig (16) Total average number of CLM stages and infested leaflets during the period of the study on hasna.

4.2. Seasonal abundance of *Semielacher petiolatus*: -

4.2.1. On lemon Citrus Limon (L)Osbeck

The population of *Semielacher petiolatus* in Fig (17) recorded four peaks of abundance (54, 37, 66. And 49 individuals/100infested leaves) appeared on 1st of September, 20th of October, 24th of November, 1st of December.

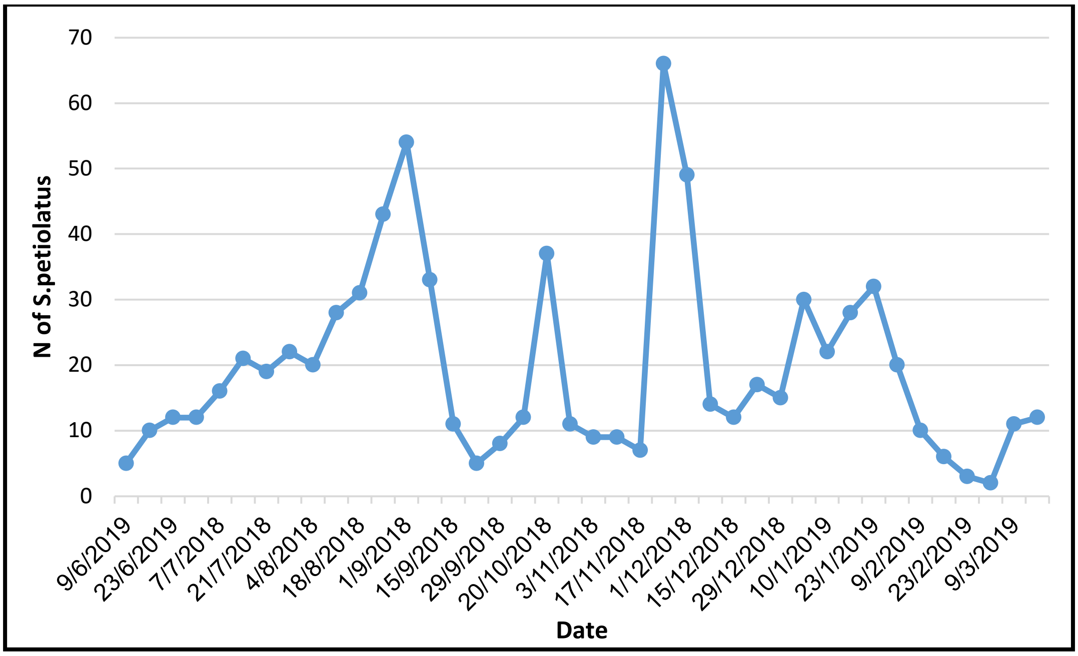

Fig (17) Population abundance of *S. petiolatus* in lemon during season 2018-2019.

4.2.2. On Abu sora, Washington navel (*Citrus sinensis osbeck*)

As presented in Fig (18) on Abu sora Washington navel the population has recorded four peaks of abundance (52, 56, 49, and 66 individuals /100infested leaves) appeared on the 7th of July, 14th of July, 1st of September, and 8th of September respectively.

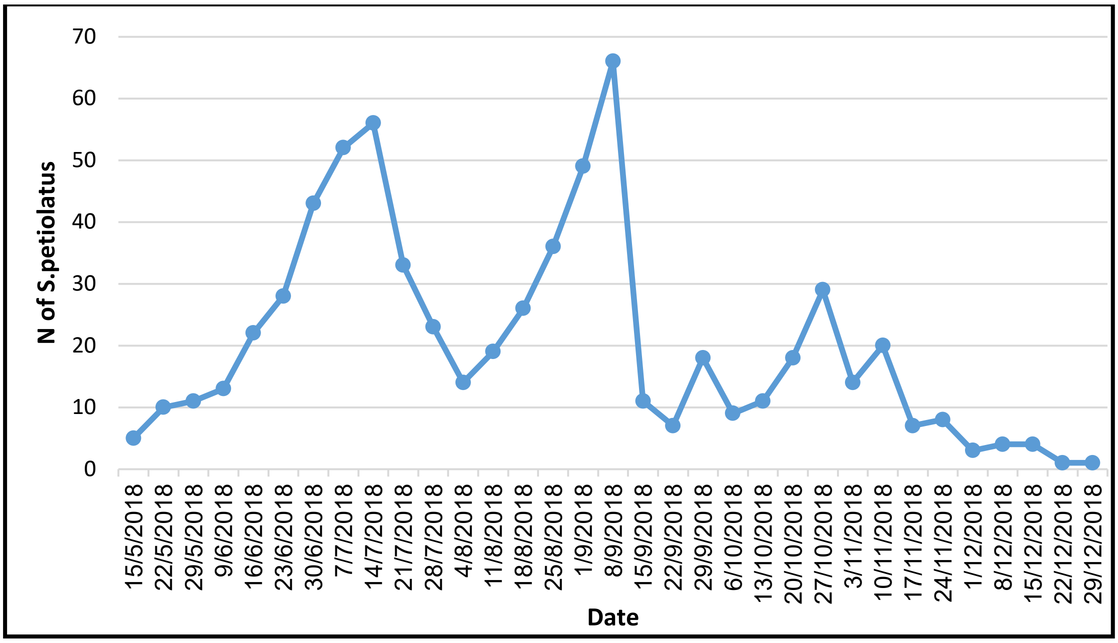

Fig (18) Population abundance of *S. petiolatus* in Abu sora Washington navel during season 2018-2019

4.2.3. on Tarocco orange (*Citrus sinensis*).

As presented in Fig (19) the population of *S. petiolatus* recorded five peaks of abundance (29, 48, 42, 59, and 38 individuals /100 infested leaves) appeared on the 14th of July, 4th of August, 18th of August, 25th of August, and 1st of September.

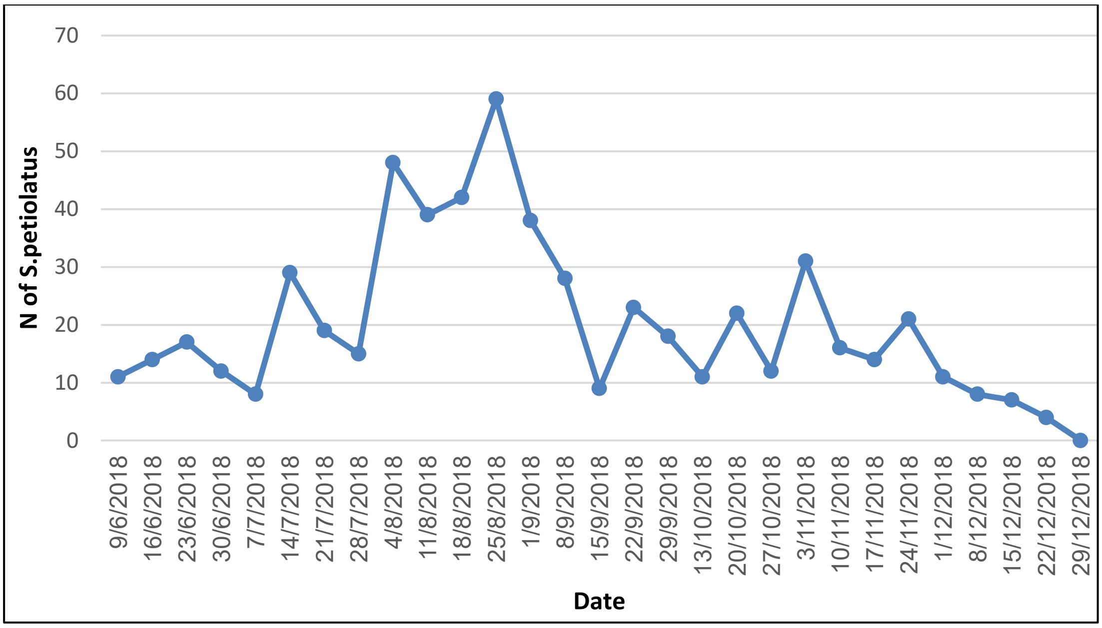

Fig (19) Population abundance of *S. petiolatus* in Tarocco orange during season 2018-2019

4.2.4. In Hasna (Citrus sinensis)

As presented in Fig (20) the population of *S. petiolatus* recorded three peaks of abundance (29, 49, and 20 individuals /100 infested leaves) appeared on the 11th of August, 1st of September, and 20th of October.

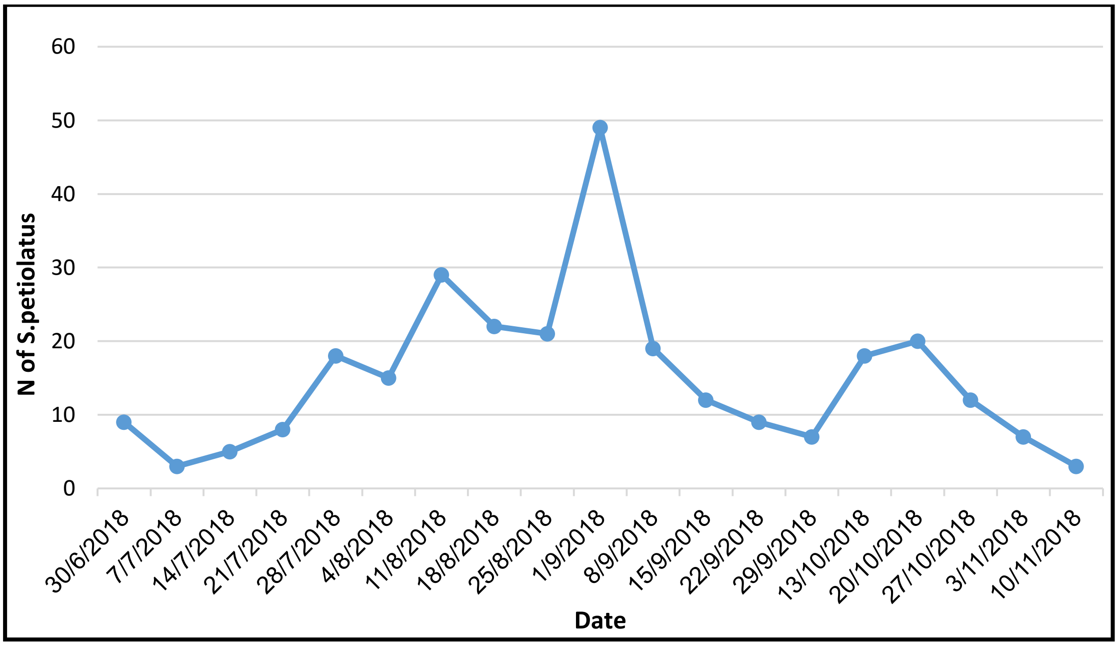

Fig (20) Population abundance of *S. petiolatus* in Hasna *(Citrus sinensis)* during season 2018-2019

4.3. Effect of mean temperature and relative humidity in the population of *P. citrella* on Abusora Washington navel.

Data in Fig (21) and Table (5) showed positive represented correlations between the population of *P. citrella* on Abusora Washington navel and mean temperature with an r-value (0.74) on the other hand, a negative correlation value was calculated between the population of *P. citrella* and relative humidity (-0.22) in Fig (22) and Table (5).

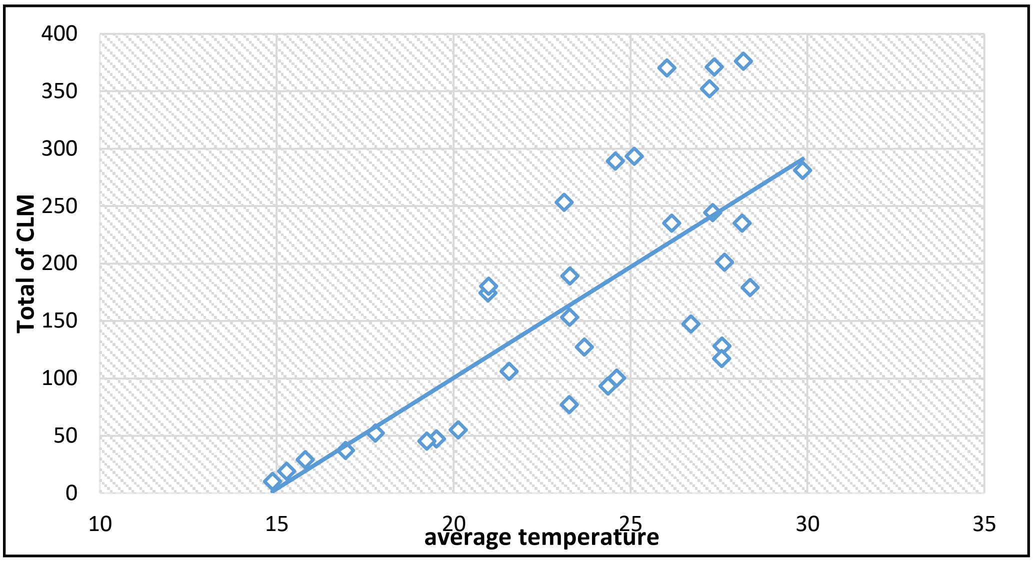

Fig (21): The liner regression showing the relation between mean temperature and population of *P. citrella* in Abusora Washington navel

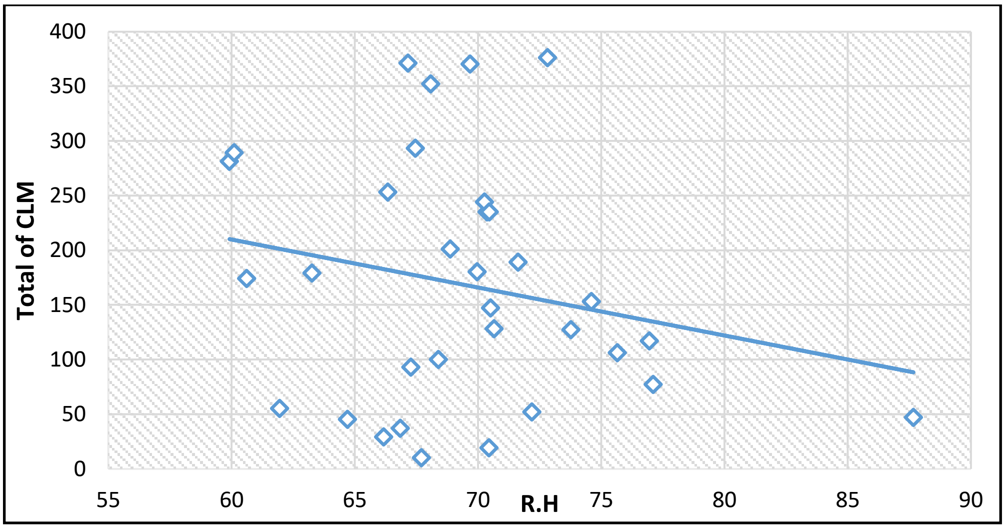

Fig (22): The liner regression showing the relation between relative humidity and population of *P. citrella* in Abusora Washington navel

Table (5): Linear regression equations and r values for population of *P. citrella* and temperature and relative humidity during 2018-2019 seasons on Abu sora, Washington navel.

The species	Mean temperature (C)		Mean R.H	
	Regression equation	r	Regression equations	r
Washington navel	Y=19.32x - 286.09	0.74	Y= - 4.39 x + 473.31	- 0.22

4.4. Effect of mean temperature and relative humidity on the population of *S. petiolatus* On Abusora Washington navel.

Data represented in Fig (23) and table (6) showed positive correlations between the population of *S. petiolatus* on Abusora and mean temperature with r value (0.69) on the other hand, a negative correlation value was calculated between the population of *S. petiolatus* and relative humidity (-0.11) in Fig (24) and table (6).

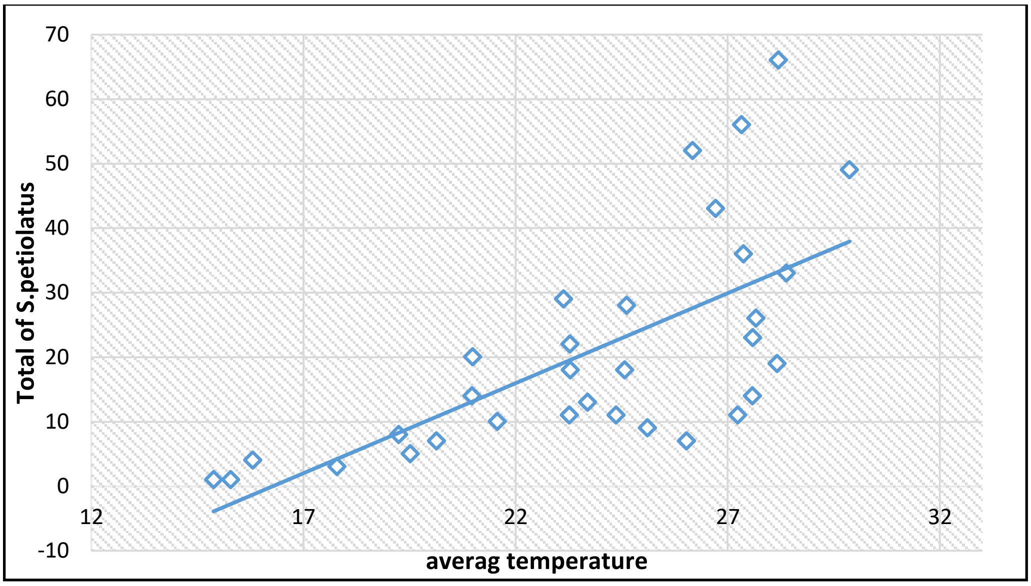

Fig (23): The liner regression showing the relation between mean temperature and population of *S. petiolatus* in Abusora, Washington navel.

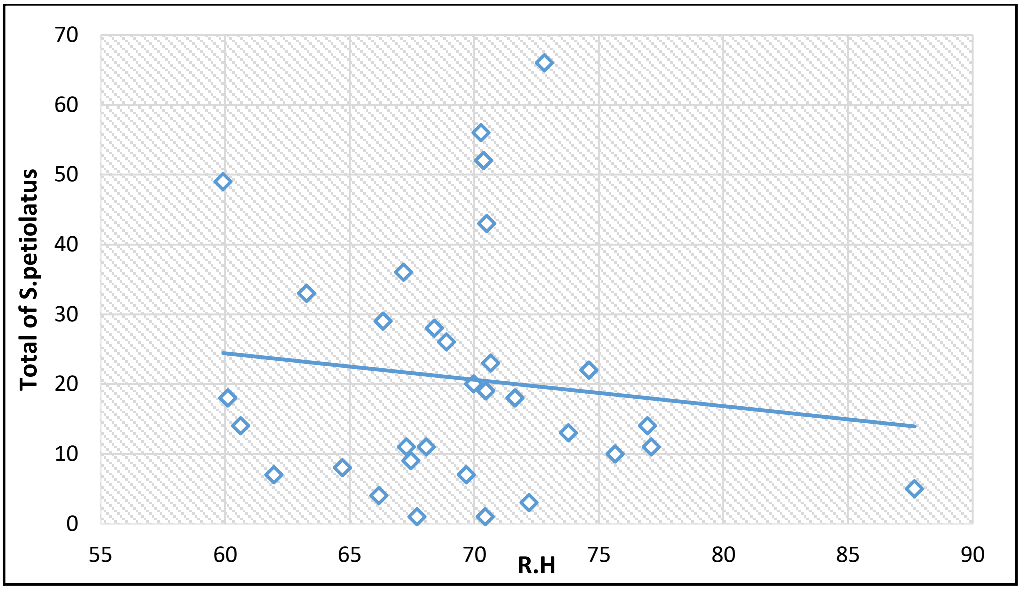

Fig (24): The liner regression showing the relation between relative humidity and population of *S. petiolatus* in Abusora, Washington navel.

Table (6): Linear regression equations and r values for population of *S. petiolatus* and temperature and relative humidity during 2018-2019 seasons on Abu sora, Washington navel.

The species	Mean temperature (C)		Mean R.H	
	Regression equation	r	Regression equation	r
Washington navel	Y=2.76 x- 44.73	0.69	Y= - 0.33 x+43.44	- 0.11

4.5. Synchronization between *S. petiolatus* and its host

Table (7) Correlation coefficient values and regression equations between *semielacher petiolatus* and *Phyllocnistis citrella* populations on four host plants.

Host plant	Semielacher petiolatus	
	Regression equations	r
Lemon	Y=0.1159x+3.1876	0.68
Washington navel	Y=0.0849x+6.2077	0.55
Tarocco	Y=0.1062x+3.4878	0.65
Hasna	Y=0.104x+2.8385	0.64

As presented in table (7) Synchronization between the ectoparasitoid *S. petiolatus* and its insect host, the citrus leaf miner *P. citrella*, was correlated more with host density in citrus lemon with (r =

0.68) fig (25), in Washington navel with (r = 0.55) fig (26), in Tarocco orange (r = 0.65) fig (27) and Hasna blood orange with (r = 0.64) fig (28).

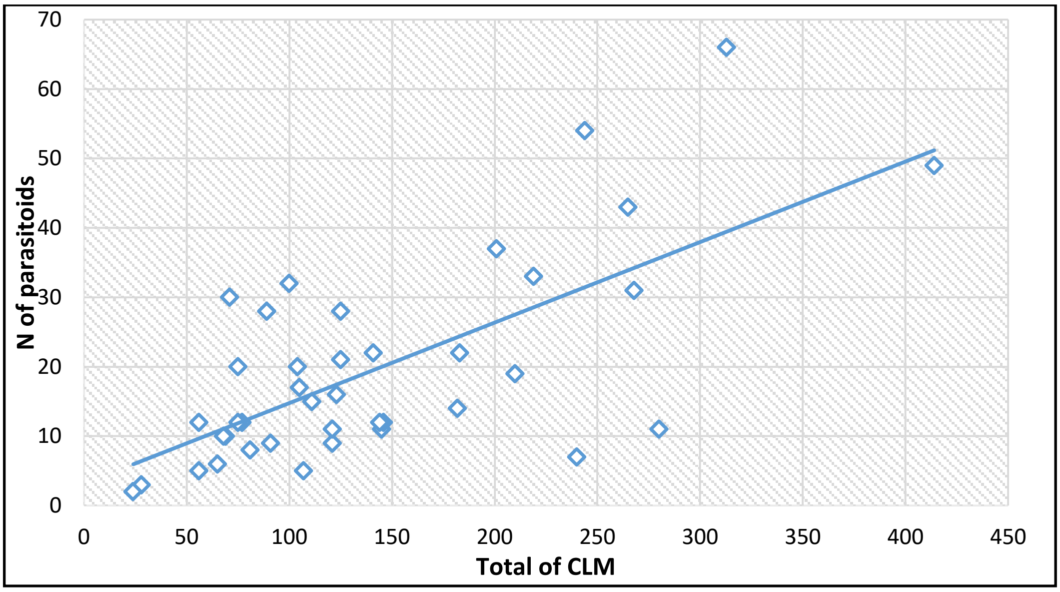

Fig (25) Synchronization between *S. petiolatus* and *P. citrella* in lemon *Citrus limon (L)Osbeck*

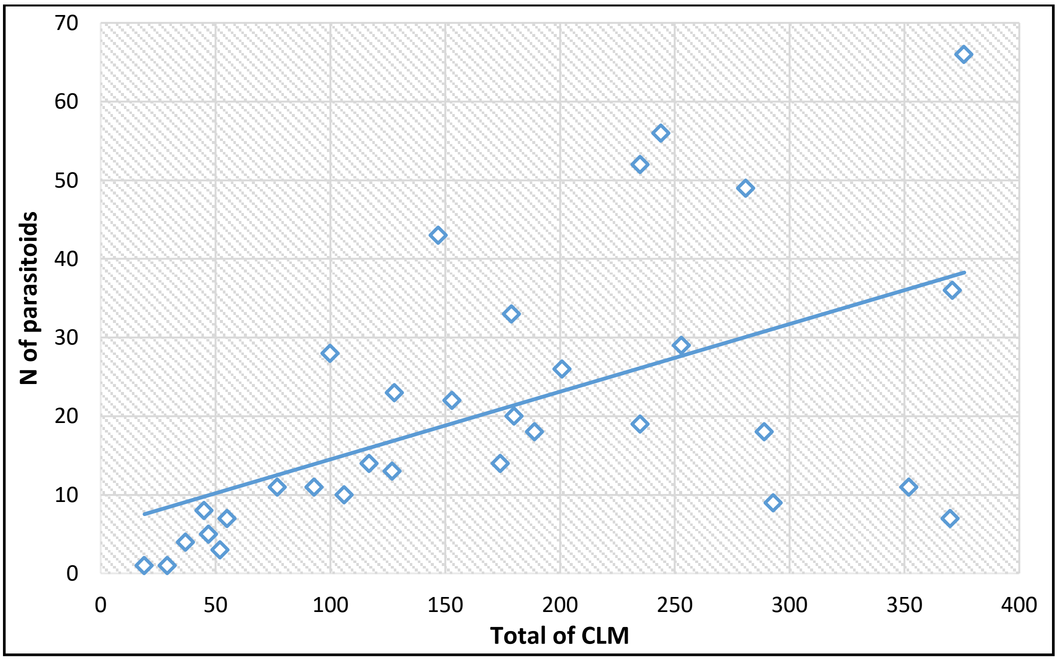

Fig (26) Synchronization between *S. petiolatus* and *P. citrella* in Washington navel (*Citrus sinensis*)

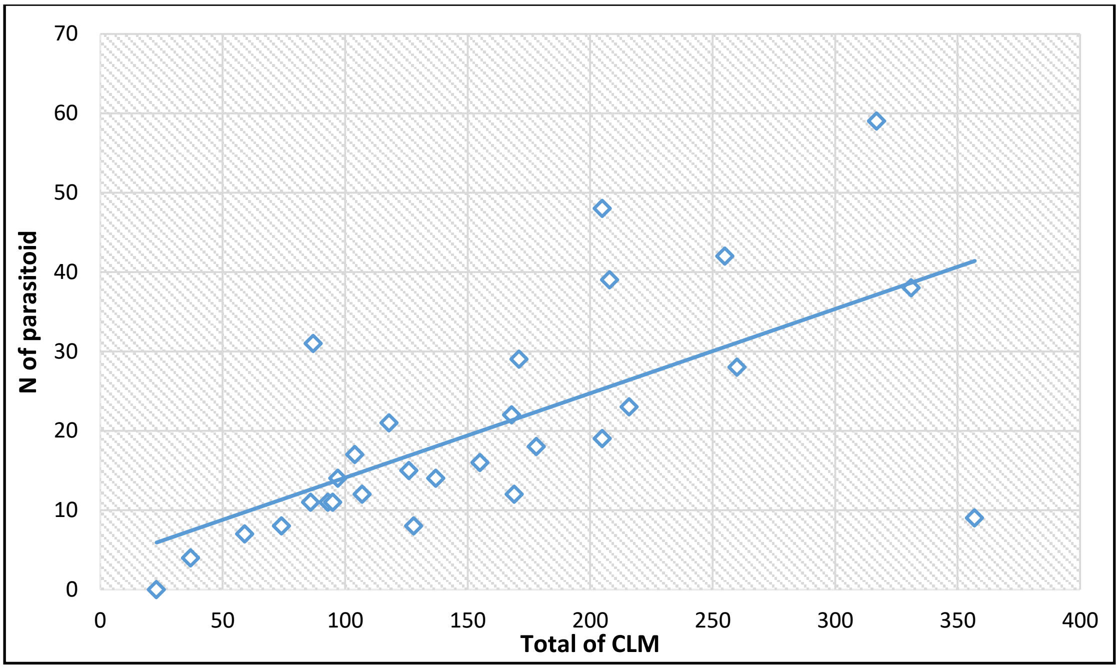

Fig (27) Synchronization between *S. petiolatus* and *P. citrella* on Tarocco (*Citrus sinensis*).

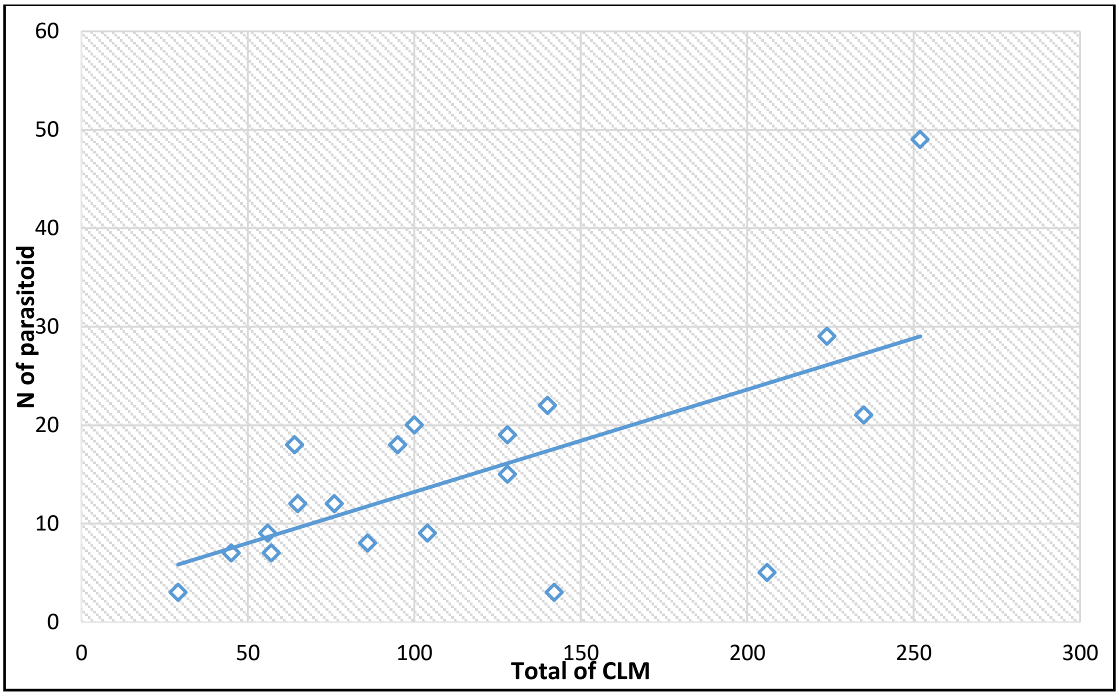

Fig (28) Synchronization between *S. petiolatus* and *P. citrella* in Hasna (*Citrus sinensis*).

4.6. Number of mines / leaflet

4.6.1. On Citrus Limon (Citrus sinensis (L)Osbeck).

As shown in fig (29) the lowest number of mines/leaflets recorded (0.6) mines/leaflets occurred on the 2nd of March 2019 while the highest number recorded (3.7) mines/leaflets occurred on the 17th of November 2018.

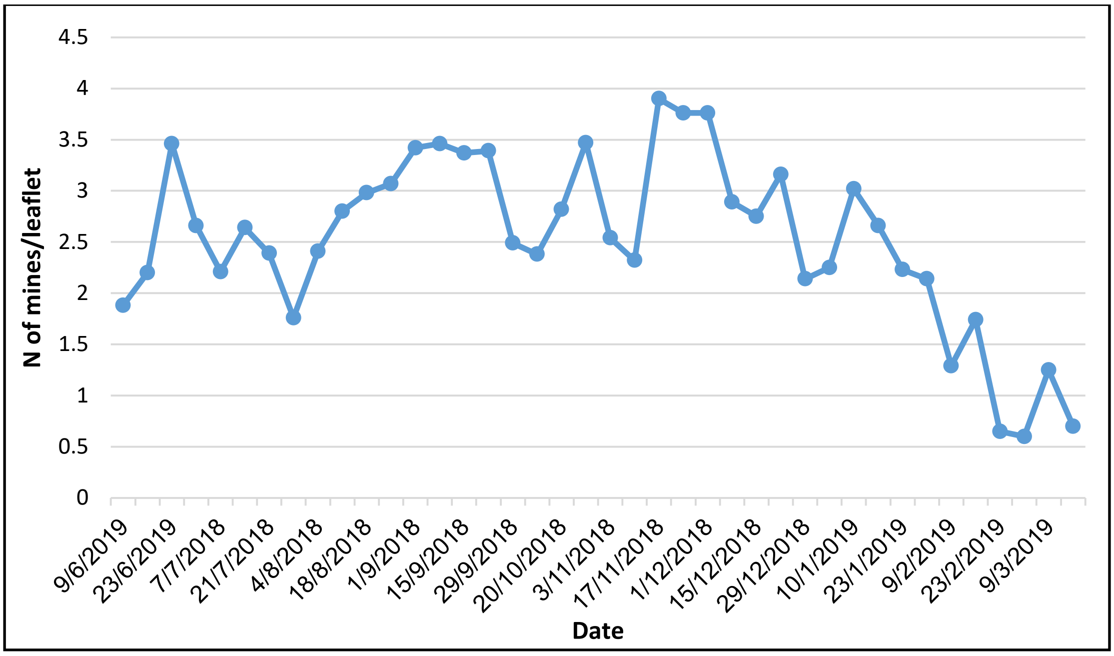

Fig (29) Number of mines/ leaflet in Lemon

4.6.2. On Abu sora, Washington navel (*Citrus sinensis*)

As shown in fig (30) the lowest number of mines/leaflets recorded (1) mines/leaflets occurred on the 29th of December 2018 while the highest number recorded (4.68) mines/leaflets occurred on the 15th of September 2018.

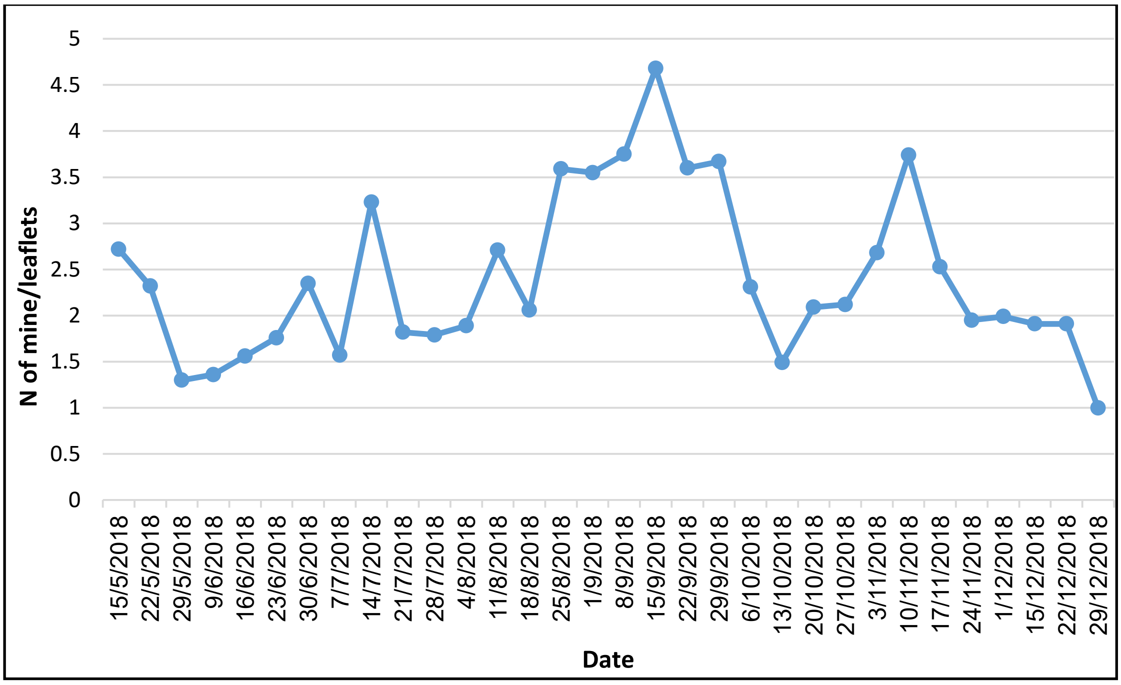

Fig (30) Number of mines/ leaflet in Abu sora Washington navel

4.6.3. On Tarocco orange (*Citrus sinensis*).

As shown in fig (31) the lowest number of mines/leaflets recorded (0.55) mines/leaflets occurred on the 29th of December 2018 while the highest number recorded (5.15) mines /100 leaflets occurred on the 15th of September 2018.

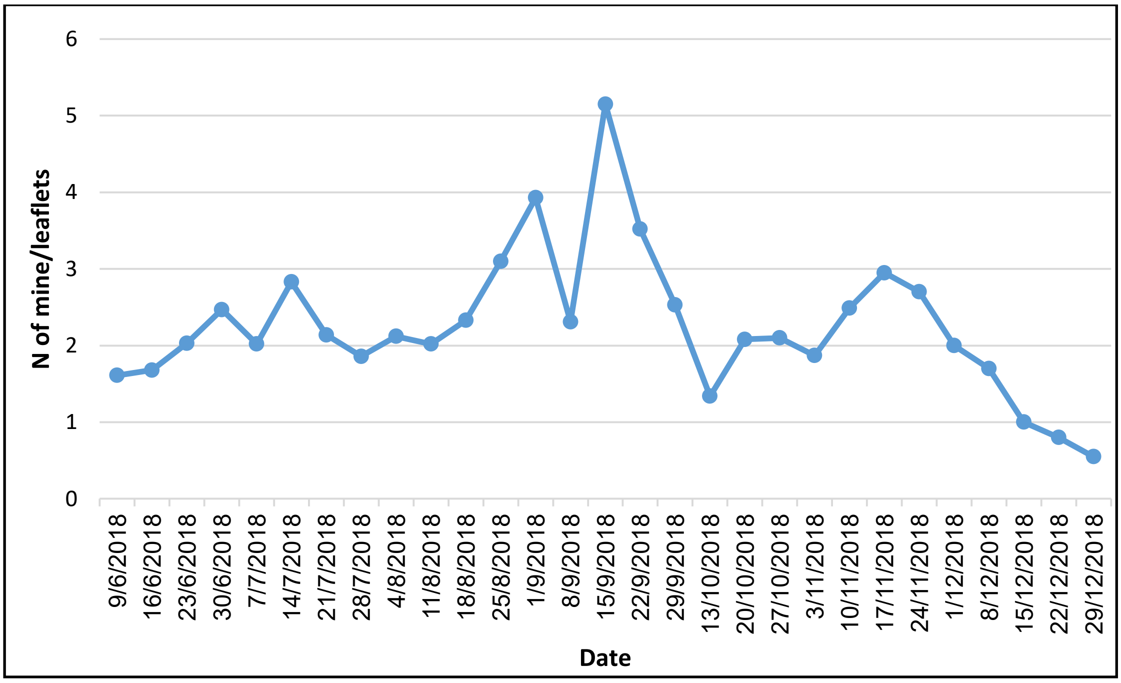

Fig (31) Number of mines/ leaflet in Tarocco orange

4.6.4. On Hasna, Blood orange

As shown in fig (32) the lowest number of mines/leaflets recorded (0.4) mines/leaflets occurred on the 10th of November 2018 while the highest number recorded (4.03) mines/leaflets occurred on the 1st of September 2018.

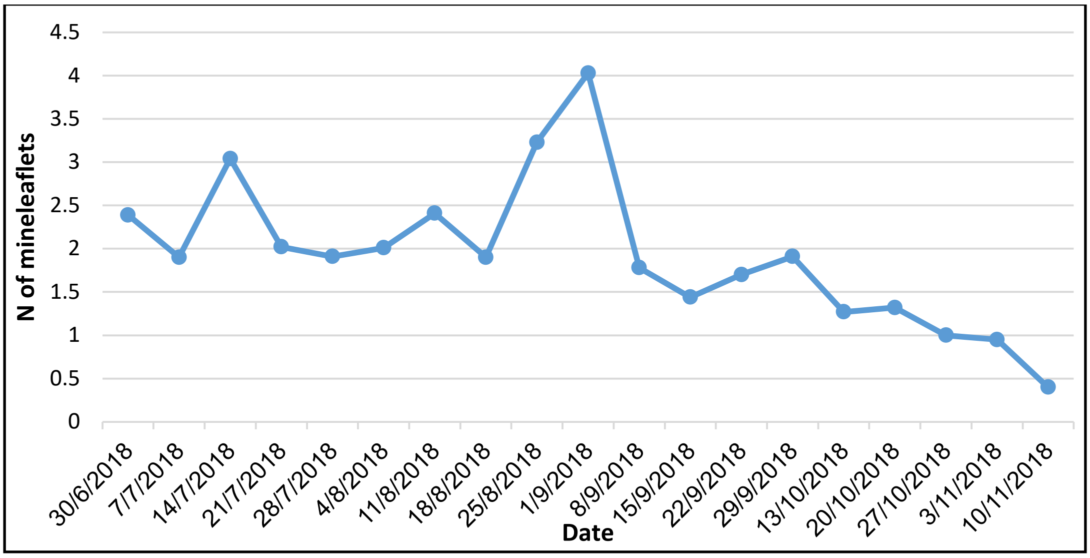

Fig (32) number of mines / leaflet in Hasna Blood orange

Data presented in Table (8) shows the total monthly average number of mines ± SD of *P. citrella* larvae in all host plants in the study, As showed results of their highest monthly average number of mines for lemon, *Citrus limon* in November (3.13 ± 0.82 mines/ 100 infested leaflets) On the other hand, was lowest monthly average numbers in March (0.85 ± 0.35 mines/100 infested leaflets) while recording the highest monthly average number of mines for Abusora Washington navel (*Citrus sinensis*) in September (3.85 ± 0.47 mines/100 infested leaflets) and lowest monthly average numbers were in December (1.74 ± 0.42 mines/100 infested leaflets). In Tarocco (*Citrus sinensis*) was the highest monthly average number of mines in September (3.49 ± 1.15 mines/100 infested leaflets) and lowest monthly average numbers was in December (1.21 ± 0.62 mines/100 infested leaflets) while recording the highest monthly average number of mines for Hasna or blood orange (*Citrus sinensis*) in August (2.39 ± 0.60 mines/100 infested leaflets) and lowest monthly average numbers was in November (0.68 ± 0.39 mines/100 infested leaflets).

Table (8) Total monthly average number of mines ± SD of *P. citrella* Larvae in all host plants during the period of the study:

months	Lemon Mean ± S.d	Abusora Mean ± S.d	Tarocco Mean ± S.d	Hasna Mean ± S.d
May	-	2.11 ± 0.73	–	–
June	2.55 ± 0.69	1.75 ± 0.43	1.95 ± 0.39	–
July	2.25 ± 0.37	2.10± 0.76	2.21 ± 0.43	2.22 ± 0.55
August	2.82 ± 0.29	2.56 ± 0.77	2.39 ± 0.49	2.39 ± 0.60
September	2.23 ± 0.41	3.85 ± 0.47	3.49 ± 1.15	2.17 ± 1.05
October	2.89 ± 0.55	2.00 ± 0.36	1.84 ± 0.43	1.20 ± 0.17
November	3.13 ± 0.82	2.73 ± 0.75	2.50 ± 0.46	0.68 ± 0.39
December	2.94 ± 0.59	1.74 ± 0.42	1.21 ± 0.62	–
January	2.46 ± 0.37	–	–	–
February	1.23 ± 0.55	–	–	–
march	0.85 ± 0.35	–	–	–

CHAPTER – V:

DISCUSSION

5. Discussion

5.1. Seasonal abundance of *P. citrella*:

As presented in the results in Tables 1 to 4 and Figures from 1 to 12 it could be concluded that the population of CLM showed 3–7 peaks of abundance on all targeted host plants, where a low number was recorded during the winter months and the beginning of spring. This can be justified by the fall in temperatures and the scarcity of young leaves. Then it recorded its highest levels in Summer and Autumn, coinciding with new citrus flushes and favorable temperature for *P. citrella* development. Similar results were obtained by **Pena *et al*. (1996)**, who found that the high peaks of the CLM population were observed during the summer (June–July) and fall (September–October) in Florida. These results also agree with the results of **Mafi and Ohbayashi. (2004)** in that the study discovered two *P. citrella* infection maximum in July and October, which were closely associated with temperatures that were ideal for growth and constant flushing of new shoots. In Sicily (Italy), **Caleca and LO Verde (1997)** report that the spring outbreak is spared from infestations of *P. citrella*, with contamination only beginning in the second half of June. **Pinto and Fucarino (2000)** report that in Sicily, summer and fall are CLM's most active periods. These results also agree with the results of **Al-Khateeb *et al*. (1999)** and **Jafari *et al.* (2000)** in that the highest density of the pest was during the summer growth period, specifically in July and August, and differs from them in the decrease in infection on the autumn growths. Through the results, it is shown that the CLM population fluctuations are affected by the high rate of a new flush as well as weather factors such as temperature and humidity, according to **Setamou *et al*. (2010)**, the new flashes which increase with increasing temperatures and sunlight activity are the most important biological factors for CLM population density, the established **Hassina *et al* (2017)** that there are two separate phases that determine the miner's. The first summer-autumn, when the weather is ideal and the leaves are tender. The second phase is the winter-spring season when there is little to no activity from miners. The drop in temperature and scarcity of the young leaves provide justification for this.

5.2. Seasonal abundance of *S. petiolatus*:

As shown in Figures 13 to 16 it could be concluded that the parasitoid *S. petiolatus* was represented in low or moderate numbers in June on four studied host plants, and then the population increased gradually, reaching its peaks during summer and autumn, especially in August and September. These results coincided with those of **(Alkhateeb *et al*., 1997; Ateyyat., 2002)** who recorded an increase in CLM density with a parasitism peaked between August and September. These results also agree with the study of **Abu Kaf *et al*. (2005)**, where the results of the statistical analysis of this study showed a high density of the parasitoid *S. petiolatus* during the summer and autumn months in Syria. These results also agree with the study of **Kalaitzaki *et al*. (2011)**, where the results showed the presence of the parasite from mid-June to early December, while it differed in its low density during the fall, according to **Gharib *et al*. (2019)**, the highest parasitism was 42.5% at the beginning of August in 2013-2014 and 19.2% at the middle of September in 2014-2015.

5.3. Effect of mean temperature and relative humidity in the population of *P. citrella* on Abusora Washington navel:

The results in Figures (17,18) and Table (5) are in parallel with those of **Mohsen. (2019)**. Statistical analysis obtained showed that the correlation between the activity of *P. citrella* population and mean temperature was positively significant. **Ahmed *et al*. (2013)** reported similar findings of a significant positive correlation between citrus leaf miner infestation with minimum temperature, and relative humidity negatively influenced CLM infestation. Contrastingly, while **Shivankar and Rao. (2003)** found a positive correlation between maximum and minimum temperatures and citrus leafminer infestation, while relative humidity showed a negative correlation. **Prabhudev (2021)** recorded A significant positive correlation between CLM incidence and maximum relative humidity (r=0.301). According to the findings **Ali and Ali. (2018)**, climatic conditions are important factors in determining the intensity of *P. citrella* incidence. Maximum and minimum temperatures had a negative association with the *P. citrella* population, although relative humidity had a positive correlation with the pest's dynamic population. Our results also agreed with **Elnagar and Soliman. (2016)**, where the positive correlation between the population of *P. citrella* and temperature, differed from them in that the relationship with relative humidity was positive.

5.4. Effect of mean temperature and relative humidity on the population of *S. petiolatus* on Abusora Washington navel:

According to the findings of this study, in Figures 19,20 and Table 6 Which can be explained by the thermal needs of the development of *S. petiolatus*. **Kalaitzaki *et al*. (2021)**, found that super parasitism occurred at 20, 25, and 30 °C, respectively so that *S. petiolatus* is much better adapted at higher temperatures typical for the Mediterranean climatic conditions during summer while its population increases are expected to be marginal at temperatures close to 20 °C. Similar results were obtained by **Braham *et al*., (2006)** who indicated a high rate of parasitism by *S. petiolatus* due to the effect of dry climatic conditions in summer and most of the autumn season. so the results of our study are in agreement with their suggestions.

Low rates of R.H. coinciding with the increase of temperature could enable parasitoid development in the winter seasons in contrast high rates of R.H. coinciding with high temperatures (in the optimal thermal range) are suitable climatic conditions for *S. petiolatus* development in the summer growing seasons. It could be concluded that the positive correlation relationship that was estimated between mean temperature and *S. petiolatus* density is due to the increase of developmental rate by the temperature increase the results are in line with those of **Kalaitzaki *et al*., (2011)**.

5.5. Synchronization between *S. petiolatus* and its host:

Data obtained in the current represented the synchronization between the ectoparasitoid *S. petiolatus* and insect host *P. citrella* was good in citrus varieties studied figure (21, 22, 23, and 24) and Table (7). where data shows, that *S. petiolatus* density did not differ significantly between the citrus varieties studied, this indicates that the parasitoid can be effective in all citrus varieties studied, These results agree with **Kalaitzaki *et al*. (2011)**, that showed through earlier field studies conducted in Chania showed that the percent of parasitism of *P. citrella* by *S. petiolatus* did not significantly differ between the citrus varieties studied, concluded results that different citrus varieties don't affect the lower survival threshold of *S. petiolatus*, this is caused by the fact that *S. petiolatus* evolution is significantly influenced by the availability of its host *P. citrella* through host feeding, these results agreed with **(Arshad., 2021)**

where the results showed that there was no significant difference in *P. citrella* incidence of Sargodha, **Lim and Hoy (2005)** found that *S. petiolatus* females kill citrus leaf miner by parasitism and host feeding However, concurrent host feeding had no effect on *S. petiolatus* growth time or adult emergence, as this could be predicted to lower host quality.

5.6. Number of mines / leaflet:

A total of 100 leaves per host plant were examined for mines caused by leafminers. The mean number of mines was estimated by using Microsoft Excel, and the results showed in Fig (25,26,27, and 28), and Table (8) an increase in the number of mines in autumn and summer in all host plants in the study, while the number of mines decreased during the winter for both Abusora and Tarocco. at the beginning of spring in

Lemon and the late autumn for Hasna blood orange. The number of mines reflects the extent of the infection, as they are caused by feeding the larvae. Due to this, the number of mines increases in the autumn and decreases in the winter. with increasing population density. According to **Malausa, (1997)**, the reason for this insect's inactivity during this time is that the adult density is low throughout the winter. In addition, the indirect effects of climate-caused stressed leaves would be harmful to the larvae. **Mingdo *et al*. (1989)** and **Huang *et al*. (1989)** reported that the primary cause of death in winter and early spring of CLM generations was the lack of water in the leaves. But, according to **Deng and Garrido (1999)**, the amount of water in the leaves did not affect how many CLM larvae died. They also argued that the cold winter weather and the fact that there weren't any new leaves growing were the main reasons why the CLM disappearing during that time. where the number of mines reflects the extent of the infection, caused by feeding the larvae. Due to this, the number of mines increases in the autumn and decreases in the winter. with increasing population density. these results agree with the study of **Liu and Beattie. (2008)** where the results showed the mean number of mines per tree per month was very low during the cooler months (November to March) while large numbers of mines were detected in May to July, and the numbers of mines were significantly higher in June. This is also consistent with the results of **Rahman., *et al* (2005)** found area of leaf infestation was observed in April but it was reduced to the minimum in July. An increase in the area of leaf infestation was again observed in August, which reached a peak in September. These results are in agreement with those of **Legaspi *et al*., (2001)** and **Ahmed *et al*., (2013)**. who stated that the percentage of harm caused by CLM peaked in September and then decreased between January and March. **Powell., *et al* (2007)** reported that May to July saw the discovery of a large number of mines, which seriously damaged the young leaves where they were found. The mean number of mines per tree per month was very low during the cooler months (November to March) in all study years. also

Kumar., *et al* (2023) found that there were 1.81 to 9.59 live mines per shoot in the citrus leaf miner population. The months of August, December, and January saw the pest's highest activity. April and May were the months with the lowest incidence.

The results of the present study showed differences in the monthly mean number of mines between selected citrus cultivars Variations in leaf thickness and specific anatomical changes could be the cause of the damage level variation **(Matthews *et al*., 2007)** These results agree with the study of **Arshad *et al*.** (**2019)** where the results showed the mine area generated by CLM larvae was significantly different on citrus cultivars.

CHAPTER – VI:

CONCLUSIONS & RECOMMENDATIONS

6-1. Conclusions

1. Based on the obtained results, it can be concluded that the population dynamics of *P. citrella* throughout the study in the Surman region were higher in the summer and autumn seasons than in the winter and spring seasons. It started in July and reached a population peak in September and October and then started to decline in the winter and spring months.
2. Climatic conditions were found to be one the important factors in determining the intensity of *P. citrella* incidence. There was a positive correlation between mean temperatures and the population of *P. citrella*, but a negative correlation between relative humidity and the population dynamic of the pest, and as such, further investigation is required.
3. The present results showed parasitism by *S. petiolatus* is an important factor in suppressing that CLM population and should be encouraged in citrus orchards. Mass rearing of *S. petiolatus* for inundative release should be evaluated for better management of CLM. Our finding highlights the role of density natural enemies against CLM which could be helpful in pest management programs. Further studies should be conducted on the assessment of natural mortality factors acting on the CLM population in Libyan citrus.

6-2. Recommendations:

1. Future research on the leaf miner's natural enemies and population dynamics will be critical for tracking the insect's development in citrus orchards and highlighting the roles that the most prevalent and effective regulatory factors play across Libya.
2. Spring growth, which is the most important stage in the development of citrus trees, has a lower prevalence of pests, thus it is imperative to encourage it, particularly in young plants.
3. Joint efforts are needed to develop an integrated pest management program combining biological control techniques, natural enemies, bio-insecticides, and biotechnological technologies to reduce insect populations.

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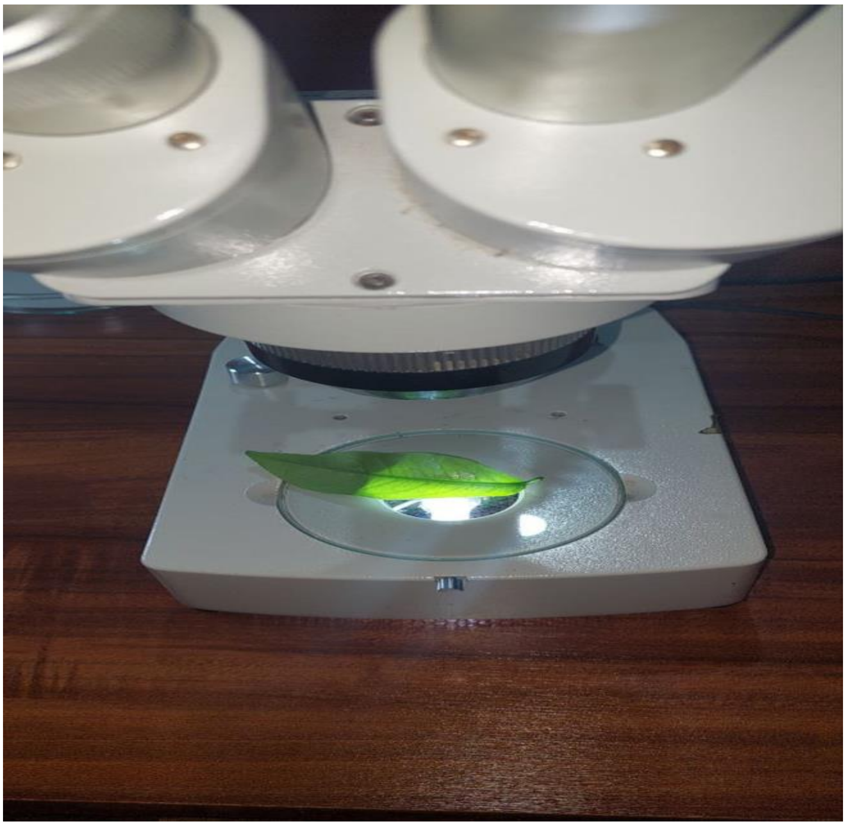
Appendix of photos

The map of the orchard from Google Earth

The orchard used to collect samples

Harvesting citrus leaves in the orchard



A citrus leaf under a binocular stereo-microscope

Citrus leaves infected with citrus leaf miner

citrus leafminer larvae and pre pupa

citrus leaf miner pupa

الملخص:

هدفت الدراسة الحالية إلى معرفة مدى وفرة أعداد حشرة حفار أوراق الحمضيات والطفيليات المرتبطة بها في أربعة نباتات مضيفة. تم جمع 100 ورقة مصابة من كل نبات مضيف. تم حفظ العينات في أكياس بلاستيكية ونقلها للفحص في المختبر.

أظهرت النتائج أن يرقات P. citrella سجلت أربع قمم للوفرة على الليمون حدثت في 21 يوليو، و18 أغسطس، و17 نوفمبر، و1 ديسمبر، كما حدثت خمس قمم للوفرة على أبوسرة في 14 يوليو. 25 أغسطس، 8 سبتمبر، 22 سبتمبر و6 أكتوبر على التوالي، بينما تم تسجيل خمس قمم للوفرة في تاروكو، حدثت في 30 يونيو، 21 يوليو، 1 سبتمبر، 15 سبتمبر، و10 نوفمبر، علاوة على ذلك، سجلت أربع قمم للوفرة على الحسنة في 14 يوليو، و11 أغسطس، و25 أغسطس، و1 سبتمبر.

سجل طور قبل العذراء P. citrella أربع قمم للوفرة على الليمون حدثت في 7 يوليو، و25 أغسطس، و27 أكتوبر، و1 ديسمبر، كما حدثت ثلاث قمم للوفرة على أبو سرة في 7 يوليو. و14 يوليو و20 أكتوبر وسجل ثلاث قمم للوفرة على تاروكو حدثت في 30 يونيو و7 يوليو و20 أكتوبر، بينما سجل على الحسنة ثلاث قمم للوفرة حدثت في 7 يوليو. 11 سبتمبر، و20 أكتوبر.

سجلت عذراء P. citrella خمس قمم للوفرة على الليمون حدثت في 16 يونيو، و30 يونيو، و10 يناير، و17 يناير، و8 مارس، وسجلت خمس قمم للوفرة على أبوسرة في 22 من مارس. مايو و9 يونيو و16 يونيو و30 يونيو و7 يوليو وسجلت أربع قمم للوفرة في تاروكو حدثت في 9 يونيو و30 يونيو و20 أكتوبر و27 أكتوبر بينما سجلت ثلاثة حدثت ذروة الوفرة في الحسنة في 30 يونيو و7 يوليو و20 أكتوبر على التوالي.

تم تحديد الطفيل الخارجي Semielacher petiolatus لمهاجمة P. citrella حيث تم تسجيل أربع قمم للوفرة على الليمون في 1 سبتمبر، 20 أكتوبر، 24 نوفمبر، و1 ديسمبر وتم تسجيل أربع قمم للوفرة على أبوسرة في 1 سبتمبر، 20 أكتوبر، 24 نوفمبر، 1 ديسمبر. 7 يوليو، 14 يوليو، 1 سبتمبر، و8 سبتمبر وسجلت خمس قمم للوفرة في تاروكو في 14 يوليو، 4 أغسطس، 18 أغسطس، 25 أغسطس، و1 سبتمبر بينما سجلت ثلاث قمم حدثت غزارة على الحسنة في 11 أغسطس، 1 سبتمبر، و20 أكتوبر على التوالي.

أشار تحليل الارتباط إلى وجود علاقة موجبة بين متوسط درجة الحرارة وأعداد P. citrella بقيمة = r (0.74) و (0.69) مع S. petiolatus بينما أظهر تحليل الارتباط وجود ارتباط سلبي ومعنوي بين الرطوبة النسبية وأعداد السكان P. citrella. بقيمة = r (- 0.22) و(- 0.11) معS. petiolatus

وزارة التعليم العالي والبحث العلمي
جامعة الزاوية
إدارة الدراسات العليا والتدريب
كلية العلوم
قسم الأحياء-علم الحيوان

الوفرة العددية لحشرة صانعة الأنفاق (فيلونيستيس-سيتريلا) وطفيلياتها في بعض أنواع الحمضيات في منطقة صرمان- ليبيا

إعداد الطالبة:

أميرة موسى الشرشاري

إشراف:

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قدمت رسالة استكمالا لمتطلبات درجة الماجستير في العلوم في علم الحيوان

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