# Estimation of the infestation rate and parasitism rate of the citrus leafminer *Phyllocnistis citrella* in a Thomson orchard near a greenhouse where beneficials are reared in Boufarik-Blida (Algeria)

# Estimativa da taxa de infestação e da taxa de parasitismo do parasitoide dos citrinos *Phyllocnistis citrella* num pomar Thomson perto de uma estufa onde são criados insectos benéficos em Boufarik-Blida (Argélia)

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## ABSTRACT

The citrus leafminer *Phyllocnistis citrella* causes severe damage to young citrus shoots in Algeria. It therefore indirectly delays fruit set and consequently production and crop profitability. The endophytic lifestyle of the insect's larvae has made chemical control difficult, making the use of other control agents essential.

To this end, a study on the population dynamics of *Phyllocnistis citrella* and the incidence of the parasite was carried out for 08 months in 2021 (from January to August 2021) in the Boufarik-Blida region (Algeria) on the Thomson variety.

The main results gathered from the sampling show that the parasitism rate reached 68% and the infection rate 39%, due to the intervention of auxiliaries (*Semielacher petiolatus*, *Cirostichus phyllocnistoides*) from the greenhouse where these species are reared near the Thomson orchard. The citrus leafminer population is declining. Parasite brokering has a positive effect on the defense

against *Phyllocnistis citrella* and represents an effective method of reinforcing biological control of this pest. In addition, the installation of mini greenhouses for the rearing of beneficials in orchards infested by citrus leafminer can be proposed as a means of reinforcing biological control. Moreover, regional containment is a healthy alternative in the context of integrated pest management, helping the ecosystem to reduce pollution. For this reason, it is worthwhile scheduling containment periods in agricultural regions.

**Keywords:** *Phyllocnistis citrella*, Thomson variety, Parasitoids, integrated pest management, Boufarik (Algeria).

**RESUMO**
A lagarta-dos-citrinos *Phyllocnistis citrella* causa graves danos aos rebentos jovens de citrinos na Argélia. Assim, atrasa indiretamente a frutificação e, consequentemente, a produção e a rentabilidade das culturas. O estilo de vida endofítico das larvas do inseto dificultou o controlo químico, tornando essencial a utilização de outros agentes de controlo.
Para o efeito, foi realizado um estudo sobre a dinâmica populacional de *Phyllocnistis citrella* e a incidência do parasita durante 08 meses em 2021 (de janeiro a agosto de 2021) na região de Boufarik-Blida (Argélia) na variedade Thomson. Os principais resultados obtidos a partir da amostragem mostram que a taxa de parasitismo atingiu 68% e a taxa de infeção 39%, devido à intervenção de auxiliares (*Semielacher petiolatus*, *Cirostichus phyllocnistoides*) da estufa onde estas espécies são criadas perto do pomar Thomson. A população de cigarrinhas dos citrinos está a diminuir. A mediação de parasitas tem um efeito positivo na defesa contra *Phyllocnistis citrella* e representa um método eficaz de reforço do controlo biológico desta praga. Além disso, a instalação de mini-estufas para a criação de organismos benéficos em pomares infestados pela larva-minadora dos citrinos pode ser proposta como um meio de reforçar o controlo biológico. Além disso, o confinamento regional é uma alternativa saudável no contexto do manejo integrado de pragas, ajudando o ecossistema a reduzir a poluição. Por isso, vale a pena programar períodos de confinamento em regiões agrícolas.

**Palavras-chave:** *Phyllocnistis citrella*, variedade Thomson, parasitóides, gestão integrada de pragas, Boufarik (Argélia).

## 1 INTRODUCTION

Citrus fruits are of great importance to the economic and social development of producing countries worldwide. They constitute export products and processing into various derivatives such as juices, jams, essences, as they can be a source of employment (LOUSSERT, 1989; KHECHNA, 2011; KHECHNA et al., 2017; MAHMOUDI et al., 2017).

Algeria is one of the leading citrus-producing countries in the Mediterranean region. The Algerian citrus sector plays a key role in economic terms (KHECHNA, 2018). After enjoying a growing boom, Algerian citrus has been experiencing a considerable decline in production and fruit quality in recent years. Among the causes of this decline, diseases and pests play a key role (BERKANI, 1995 ; BERKANI et al., 1996 ; BOULFEKHAR-RAMDANI, 1998 and BERKANI, 2003). These include *Phyllocnistis citrella* STAINTON (Lepidoptera: Gracillariidae) of Asian origin, which has recently appeared in the Mediterranean basin, causing damage to leaves that

reduces photosynthetic activity. The importance of the citrus leafminer as a pest is linked to several factors. This phytophagous pest attacks young, tender citrus leaves. Its outbreak can lead to defoliation of trees and arrested shoot development, while heavy attacks can significantly reduce the photosynthetic activity of infested leaves (SAHARAOUI, 1997; SAHARAOUI et al., 2001; KHECHNA et al., 2017; KHFIF, 2022). The larvae feed on the epidermal cells and dig silvery, sinuous mines along the leaves, causing them to curl up (SMITH et al., 1997). One larva destroys 1 to 7 cm of leaf surface (SAHARAOUI et al., 2001).

SOLTANI (2013), shows that the excessive and inappropriate use of conventional pesticides (broad spectrum of action, low selectivity) against pests has disrupted the natural balance of the agricultural ecosystem (including an effect on parasitoids and predators, secondary pest infestations and the phenomenon of pest resistance). In addition, they present serious health risks for farmers and negative repercussions for the environment (contaminating air, soil and groundwater) (SOLTANI, 2013). Many countries infested by this pest have opted in particular for the introduction and acclimatization of entomophagous species, whose real effectiveness sometimes remains controversial (ARGOV and ROSSLER, (1998) ; SCHAUFF et al. (1998) ; FAZEKAS (2023) ; MIRANDA QUITIAQUEZ et al. (2023) ; ULLAH et al. (2023)).

In Algeria, apart from the species introduced in 1996 by the Institut National de Protection des Végétaux, only three parasitoids of P. citrella were known to date (SAHARAOUI et al., 2001). In 2011, the Institut National de Protection des Végétaux launched a program to revive the breeding of allochthonous Semielacher petiolatus and Citrostichus phyllocnistoïdes beneficials, and undertook releases for the implementation of biological control of the citrus leafminer *Phyllocnistis citrella*.

Indeed, the use of biological control with other control techniques can be a solution for integrated protection against pests (KHFIF, 2022 and BENDOUMIA, 2018). According to the latter author, biological control plays a vital role in reducing the population of this leafminer, and its success depends on continuous monitoring of orchards and the populations actually present in order to intervene at the right time.

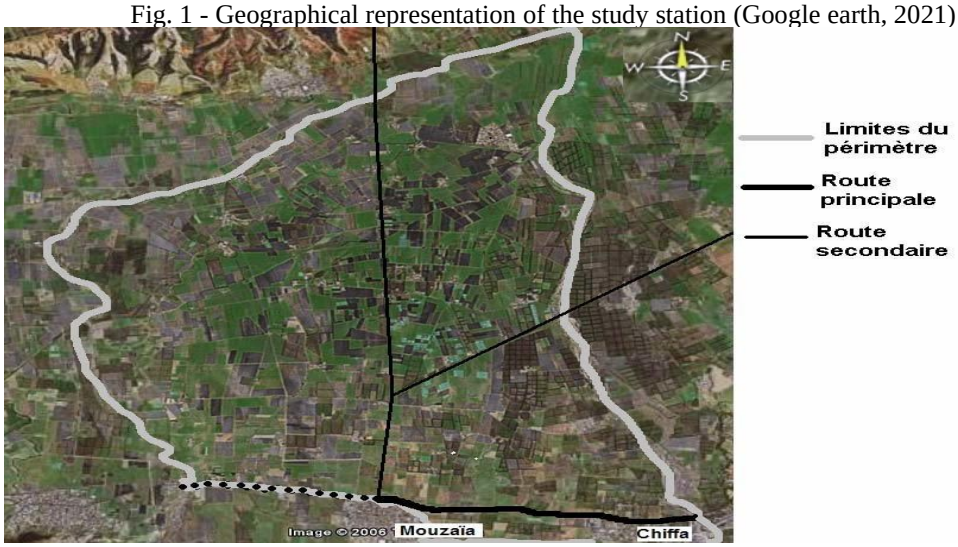
The aim of the present study is to estimate the infestation and parasitism rates of the citrus leafminer *Phyllocnistis citrella* in a Thomson (*Citrus sinensis* L.) orchard near a greenhouse where beneficials are reared in Boufarik (Algeria). The aim of this work is to identify the beneficials of this pest, which could play an important role in enhancing biological control of this citrus variety. The aim is also to minimize the use of pesticides. Pesticides cause numerous problems and illnesses through their residues in the human body and throughout the ecosystem. Furthermore, the sampling

period of the present study was carried out during the national containment as a preventive measure against the Corona virus "Covid-19" pandemic.

## 2 MATERIALS AND METHODS

The sampling of the present work was carried out in the regional plant protection station (Boufarik-Blida) (36° 59′ 49″ 42 N., 2° 91′ 98″ 26.7 L.) of the Mitidja plain in Algeria (Fig. 1). It covers an area of 3 ha and is bounded to the north, west and south by citrus orchards and to the east by the freeway. The station is located at the edge of the town of Boufarik, in a semi-urban zone, on the edge of the road leading to Tessala El Merdja. Its work is technical and administrative.

Fig. 1 - Geographical representation of the study station (Google earth, 2021)




The citrus variety chosen for this study was Thomson. This is because the trees in this orchard are in a clean state of health, "no pesticides are used in this orchard". Moreover, this orchard is located next to a greenhouse where citrus leafminer parasitoids are reared, notably Semielacher pesiolatus and Cirostichus phyllocnistoides.

It should be noted that the sampling in the present study was carried out during national containment as a preventive measure against the Corona virus "Covid-19" pandemic. In addition, outings were carried out several times a month for 08 successive months in 2021 (January to August) with periodic random population counts carried out to monitor variations in the infestation rate and parasitism rate of this pest. In the field, samples were taken at ground level from two trees in each of these blocks, at various cardinal points on the tree, on twigs containing young, medium or old shoots. The samples, consisting of 100 leaves for each sampling, were placed in paper bags bearing all the information needed to identify the sample, in particular the date of removal, the location, the variety and the orientation of the twigs on the tree.

In the laboratory, the different larval stages of the leafminer were identified under a binocular microscope and recorded per leaf surface. The different states of this pest - living, dead or parasitized - were also noted. In addition, nymphs of different species of beneficials were collected. It should be noted that the insect's developmental stages were precisely identified using scientific criteria reported in the literature, in particular those mentioned by BERKANI, (2003).

The technique used to exploit the results in the present study is the infestation rate and the parasitism rate. The infestation rate %= (Number of infested leaves /Total number of leaves) X 100. Parasitism rate % = (Number of parasitic individuals /Number of individuals of leafminer observed) X 100.

## 3 RESULTS AND DISCUSSION

In this section, results and discussions on *Phyllocnistis citrella* population fluctuation during the 2021 study period are discussed. This is followed by results and discussion of infestation rate estimation. Next, results and discussions of parasitism rate estimation are presented. Finally, results and discussions on the relationship between infestation rate and parasitism rate are developed.

### 3.1 RESULTS AND DISCUSSION OF *PHYLLOCNISTIS CITRELLA* POPULATION FLUCTUATION DURING THE STUDY PERIOD IN 2021

In what follows, we discuss the results of the *Phyllocnistis citrella* population fluctuation during the 2021 study period.

#### 3.1.1 Results of *Phyllocnistis citrella* population fluctuation during study period in 2021

Table 01 and Figure 02 show the population fluctuation of *Phyllocnistis citrella* during the 2021 study period.

Table 01 - Population dynamics of *Phyllocnistis citrella* on citrus trees in the Boufarik region (Algeria) in 2021 (Original)

| The dates | Stages and numbers | | | | | |
|---|---|---|---|---|---|---|
| | Egg | Larva 1 | Larva 2 | Larva 3 | Pre-nymph | Nymph |
| **January - May** | 0 | 0 | 0 | 0 | 0 | 0 |
| **07 June 2021** | 0 | 0 | 0 | 0 | 0 | 0 |
| **14 June 2021** | 0 | 0 | 0 | 3 | 2 | 2 |
| **21 June 2021** | 0 | 0 | 6 | 10 | 2 | 0 |
| **28 June 2021** | 0 | 2 | 0 | 2 | 4 | 6 |
| **04 July 2021** | 3 | 4 | 4 | 2 | 0 | 5 |
| **13 July 2021** | 4 | 2 | 3 | 1 | 0 | 7 |
| **21 July 2021** | 0 | 4 | 4 | 5 | 1 | 5 |
| **09 August 2021** | 0 | 3 | 3 | 1 | 1 | 1 |
| **The total** | **7** | **15** | **16** | **24** | **10** | **26** |

Fig. 02 - Fluctuation of *Phyllocnistis citrella* stages on Thomson during 2021 in Boufarik (Algeria) (Original)

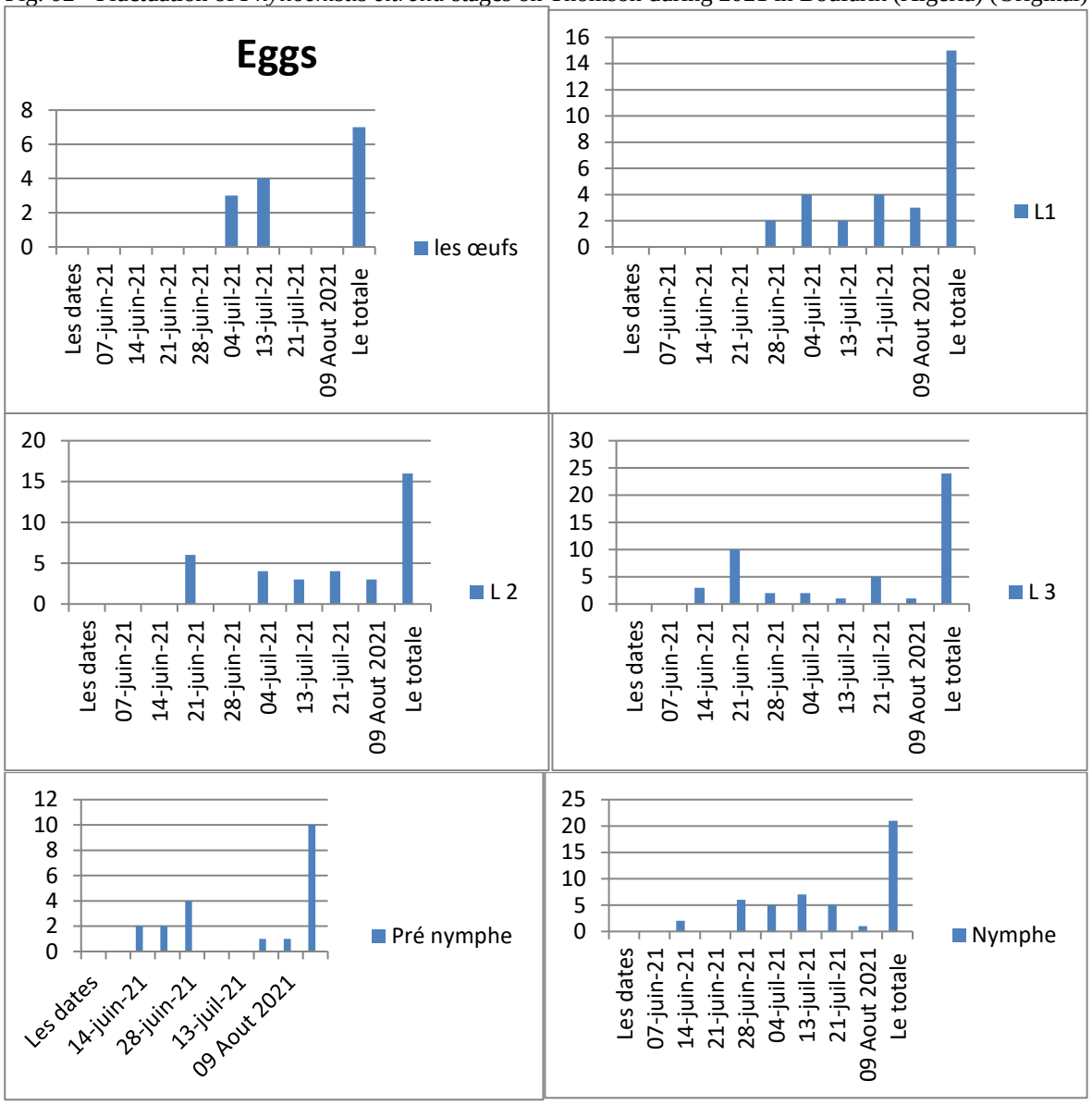



Table 1 and Figure 2 show a fluctuation in the number of *Phyllocnistis citrella* populations on the Thomson citrus variety in the Boufarik-Blida region (Algeria) during the period (January-August 2021).

The total number of eggs laid during this period was very low, fluctuating between 7 and 26 individuals. We reported a number of 15 individuals of Larva 1, 16 individuals of Larva 2, 24 individuals of Larva 3 and 10 individuals of Larva 4. The number of nymphs was 26. It should be noted that for the first few months of the study, *Phyllocnistis citrella* had zero activity or no activity at all.

This delay in Phyllocnistis citrlla establishment is due to thermal conditions from January to the first week of June, with temperatures below 18°C and a lack of food during the winter and spring. After this period, an increase in vegetation was recorded under favorable climatic conditions.

The main factors in the mortality of this pest are several, notably climatic conditions, which are essential ecological parameters in the development of any living being. Another very important

factor is the acclimatization of parasitoids. In addition, national containment as a preventive measure against the Corona virus "Covid-19" pandemic offers a cleaner climate than in previous years. In fact, this pest is rarely observed in this orchard, which is not treated with pesticides. The orchard is also close to a greenhouse where Phyllocnistis citrlla parasitoids are reared, which will minimize the establishment of this pest.

### 3.1.2 Discussion of *Phyllocnistis citrella* population fluctuation during study period in 2021

DREUX (1980), the most important parameter is temperature, as it has ecological effects on all living beings.

BERKANI et al. reported in 1996 that the number of *Phyllocnistis citrella* leafminers remains high when temperatures exceed 18° and young shoots are abundant.

Our results show that the most sensitive stage of the Phyllocnistis citrus leafminer is the third larval stage (L3) and the pre-pupal stage, confirming the findings of BOUALEM and BARKANI (2002).

KHECHNA in 2011 shows that the population of *Phyllocnistis citrella* is almost negligible or non-existent during the winter and spring periods. The summer period is the most favorable for leafminer populations, thanks to favorable temperatures and the presence of tender young leaves.

## 3.2 RESULTS AND DISCUSSION OF INFESTATION RATE ESTIMATES

In this section, the results and discussion of infestation rate estimation are developed.

### 3.2.1 Results of infestation rate estimation

Table 02 and figure 03 show the estimated infestation rate during the study period in 2021.

Fig. 03 - *Phyllocnistis citrella* infestation rate in the orchard (Original)

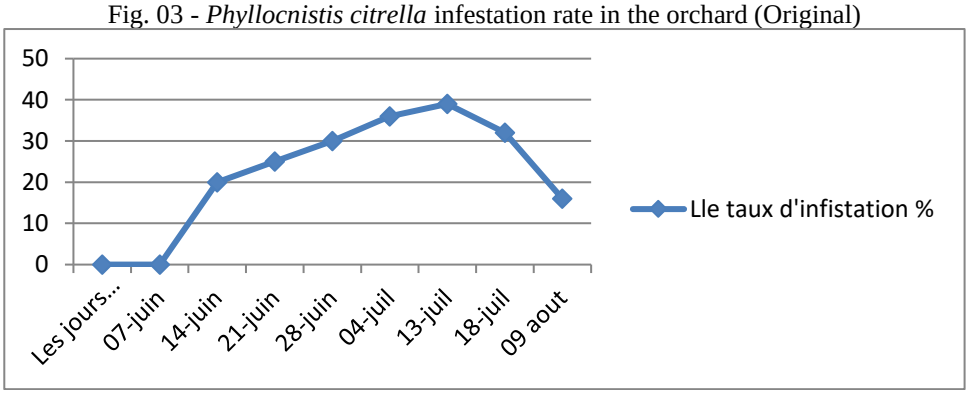


Table 02 - *Phyllocnistis citrella* infestation rate in the orchard (Original)

| Sampling days 2021 | Infestation rate % |
|---|---|
| **07 June** | 0 |
| **14 June** | 20 |
| **21 June** | 25 |
| **28 June** | 30 |
| **04 July** | 36 |
| **13 July** | 39 |
| **18 July** | 32 |
| **09 August** | 16 |

Table 2 and figure 3 show a significant increase in the infestation rate. It was recorded during the period 07 June-14 June 2021 reaching 20%. This increase continued gradually until it reached its peak in the second week of July (13-07-2021) with a rate of 39% at an average temperature of 28.9°C and humidity of 57%, forcing growers to use the best-known parasiticides (Semielacher petiolatus, Cirostichus phyllocnistoides) for complete treatment of citrus leafminer. Subsequently, the infection rate dropped from 32% on July 18 to a ratio of 16% on August 9. This variation returns to the summer push period especially in (June-August 2021) is due to climatic factors which may also be at the origin of the differences observed in pest levels, in particular temperature or the movement of *Phyllocnistis citrella* adults to neighbouring citrus trees.

### 3.2.2 Discussion of infestation rate estimates

According to KHECHNA et al. (2017), the infestation rate for the Wachinthon variety is high, with a percentage of 50% observed on July 13 and another of 60% noted on August 24 in 2011. An increase in the infestation rate can be explained by the appearance of the second sap burst in the second dekad of June. But from the third sap burst onwards, the infestation rate declines. In 2012, she reported two peaks in plot 1. The first occurred on July 18, 2012 with a rate of 35% and the second, of lesser importance, was reported on September 12, 2012 with a rate of 20%. For plot 2, three peaks of almost equal importance were noted. The first was observed on July 18, 2012 with a rate of 37%, the second peak on (August 15 of the same year with a rate of 35% and the third peak on September 12 with a rate of 38%.

## 3.3 RESULTS AND DISCUSSION OF PARASITISM RATE ESTIMATION

In this section, the results and discussion of parasitism rate estimation are presented.

### 3.3.1 Results of parasitism rate estimation

Table 03 and Figure 04 show the estimated rate of parasitism during the study period in 2021.

Table 03 - *Phyllocnistis citrella* parasitism rate in the orchard (Original)

| Sampling days 2021 | Parasitism rate % |
|---|---|
| **07 June** | 0 |
| **14 June** | 35 |
| **21 June** | 68 |
| **28 June** | 27 |
| **04 July** | 50 |
| **13 July** | 41 |
| **21 July** | 59 |
| **09 August** | 56 |

Fig. 04 - *Phyllocnistis citrella* parasitism rate in the orchard (Original)

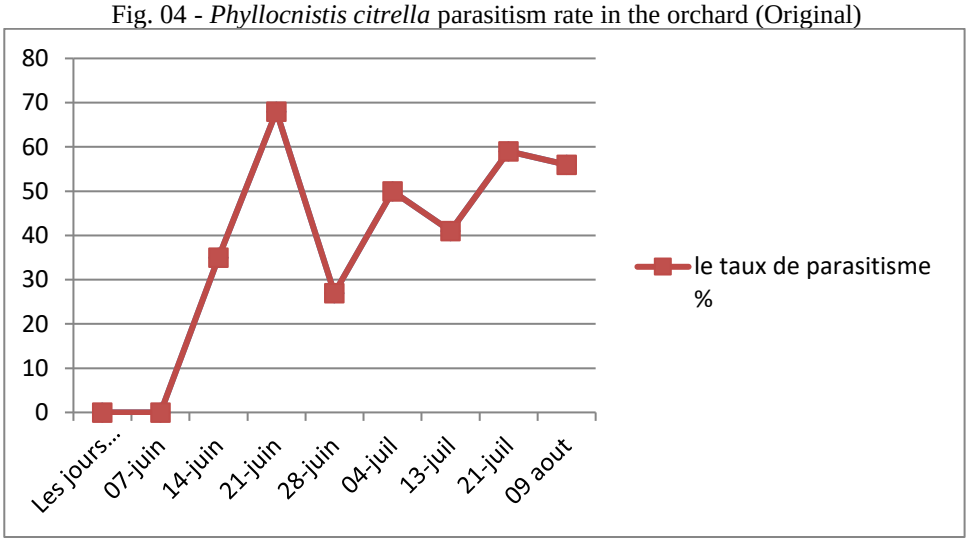



Table 3 and Fig. 4 show a significant increase in parasitism rates, reaching two main peaks, the first with 68% on June 21, 2021 and the second peak on July 21, 2021 with 72%. Thereafter, the parasitism rates fluctuate. This underlines the importance of auxiliary activity in this orchard against citrus leafminer. Near Thomson's orchard is a greenhouse where citrus leafminer parasitoids (Semielacher petiolatus, Cirostichus phyllocnistoides) are reared. In addition, this orchard is not treated with pesticides, which encourages the multiplication of many beneficials.

### 3.3.2 Discussion of parasitism rate estimates

According to FAURIE et al (1980 and 2012), chemical properties can affect the biological properties of P. citrella to some extent. Indeed, host plant quality is the key to determining the fertility of insect herbivores. The quality of the host plant also affects the insect's reproductive strategy, such as the choice of oviposition sites, which determines the fate of the larvae.

KHECHNA et al (2017), show that the first rate of parasitism is noted on June 29, 2011 with a rate of 15% and the second with a rate of 20% reported on July 27. These parasitism percentages are recorded after the first release of citrus leafminer parasitoids (Semielacher petiolatus and Citrostichus phyllocnistoides), which was carried out after the appearance of the second sap burst.

3.4 RESULTS AND DISCUSSION ON THE RELATIONSHIP BETWEEN INFESTATION RATE AND PARASITISM RATE PARASITISM

The results and discussion of the relationship between infestation rate and parasitism rate are then developed.

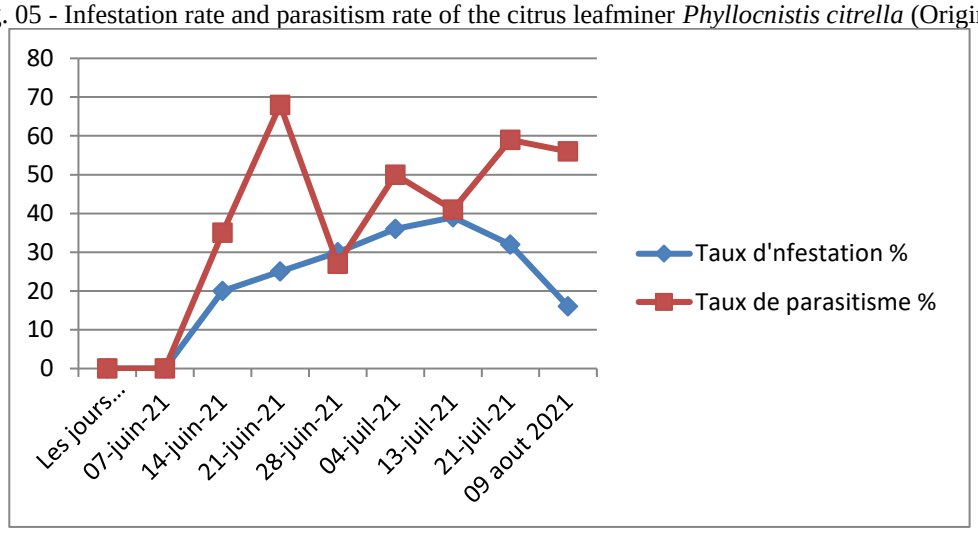
### 3.4.1 Results of the relationship between infestation rate and parasitism rate

Table 04 and Figure 05 show the relationship between infestation rate and parasitism rate during the study period in 2021.

Table 04 - Infestation rate and parasitism of the citrus leafminer *Phyllocnistis citrella* (Original)

| Sampling days | Infestation rate % | Parasitism rate % |
|---|---|---|
| **07 June 2021** | 0 | 0 |
| **14 June 2021** | 20 | 35 |
| **21 June 2021** | 25 | 68 |
| **28 June 2021** | 30 | 27 |
| **04 July 2021** | 36 | 50 |
| **13 July 2021** | 39 | 41 |
| **21 July 2021** | 32 | 59 |
| **09 August 2021** | 16 | 56 |

Fig. 05 - Infestation rate and parasitism rate of the citrus leafminer *Phyllocnistis citrella* (Original)




During the January-August 2021 study period, the results obtained concerning the fluctuating relationship between infestation rate and parasitism rate, are collated in Table 4 and Figure 5.

The parasitism rate is the most important in relation to the infestation rate, increasing and reaching a maximum of 68% in summer sap shoots on June 21, 2021.

According to the results obtained in Table 4 and Figure 5, it should be noted that the parasitism rate is higher than the infestation rate. The relationship between the rate of infestation

and the rate of parasitism is increasing, but the value of the rate of parasitism exceeds that of infestation.

The significant relationship between the rate of parasitism and the rate of infestation is due to several factors, in particular thermal variations. This can be explained by the proper conduct and success of the biological control method involving the release of parasitoids.

In addition, the increase in the rate of parasitism in relation to the rate of infestation is a result of the absence of chemical treatments in the experimental orchard. This gives the trees (Thomson) the strength to fight pests naturally (high parasitism rate compared with infestation rate).

It's also important to point out that 2021 is a year of confinement in this region, which means a clean climate (not many cars, less atmospheric gas, less air pollution from factories, less pollution in general...).

### 3.4.2 Discussion of the relationship between infestation rate and parasitism rate

KHECHNA et al. (2017) in 2011 found the infestation rate to be higher than the parasitism rate. This is due to pesticide treatments. The rate of parasitism increased from August 15, 2012 compared with the infestation rate, underlining the importance of beneficial insect activity. In fact, the importance of the parasitism rate in relation to the infestation rate is due to the activity of beneficials.

## 4 CONCLUSION AND OUTLOOK

During the study period, the parasitism rate reached 68% and the infection rate 39%, due to the intervention of auxiliaries (Semielacher petiolatus, Cirostichus phyllocnistoides) from the greenhouse where these species are reared near the Thomson orchard. In addition, the orchard is characterized by the absence of chemical treatments, which favours the abundance of auxiliary fauna.

This indicates that the citrus leafminer has good parasitic activity during national containment as a preventive measure against the Corona virus "Covid-19" pandemic in 2021. In addition, we suggest installing mini greenhouses for rearing beneficials in citrus leafminer-infested orchards to reinforce biological control. At the end of the rearing period, the doors of the greenhouse will be opened so that the beneficials can leave directly for the orchard infested by *Phyllocnistis citrella*.

Moreover, regional containment is a healthy alternative in the context of integrated pest management, helping the ecosystem to reduce pollution. For this reason, it's a good idea to schedule containment periods in agricultural regions.

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