



Egyptian Journal of Plant  
Protection Research Institute

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**Effect of mean temperature and relative humidity on the population abundance of the serpentine leaf miner *Liriomyza trifolii* (Diptera: Agromyzidae) and its parasitoids *Diglyphus isaea* (Hymenoptera: Eulophidae) and *Opius pallipes* (Hymenoptera: Braconidae)**

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**ARTICLE INFO**

*Article History*

Received: 11/1 /2024

Accepted: 12 /3/2024

**Keywords**

*Liriomyza trifolii*,  
temperature, relative  
humidity, parasitoids  
and population.

**Abstract**

The present study was carried out in Surman region kidney beans and broad beans were selected as summer and winter host plants during seasons 2020/2021. Correlation values between the population of *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) and mean temperature (°C) were recorded (0.81 and -0.27) in the summer and winter seasons, respectively. A negative correlation was calculated between the population of *L. trifolii* and relative humidity (RH.) in the summer season (-0.27) and a positive correlation occurred in the winter season recording (0.19). Correlation recorded between the population of *Diglyphus isaea* (Walker) Hymenoptera: Eulophidae) and mean temperature °C recorded (0.70 and -0.1) in the summer season and winter, respectively. On the other hand, correlation values were calculated (-0.17 and 0.36) between the population of *D. isaea* and (RH.) in the summer and winter seasons respectively. Positive correlations were recorded between the population of *Opius pallipes* (Wesmeal) (Hymenoptera: Braconidae) and mean temperature °C in summer and winter seasons with r values (0.61 and 0.035). Moreover, a negative correlation was calculated between the population of *O. pallipes* and (RH.) in the summer and winter seasons with r values (-0.30, and -.19), respectively.

**Introduction**

Agromyzidae (Diptera) is a large family of phytophagous a calyptrate flies, with almost 3,000 known species around the world (ITIS, 2016). The genus *Liriomyza* is one of the richest Dipteran taxa, containing more than 300 globally distributed species. Their larvae inhabit and consume leaf mesophyll of miscellaneous plants (Kwon *et al.*, 2018). The majority of *Liriomyza* species (nearly 23 species) are polyphagous

herbivorous

attacking crops and ornamentals of various plant families (Kang *et al.*, 2009). According to Benavent-Corai *et al.* (2005), nearly 900 species in the world have their host plants, distributed among more than 140 plant families. *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) is one of the most economically important species of the genus *Liriomyza*. It is mainly distributed in African,

Asian, and Latin American countries (Morgan *et al.*, 2000).

Depending on estimates of the best, lower, and higher temperature thresholds for all life stages, temperature is a crucial abiotic factor impacting arthropod population dynamics and limiting biological activity. Moreover, temperature features might differ between species, populations, phases of development, and other ecological factors (Bybee *et al.*, 2004). Temperature has an impact on insect development, allowing for species-specific features. The synchronization between pests and natural enemies can be determined using biological factors and developmental rate models (Kontodimas *et al.*, 2004).

The oviposition behavior of *L. trifolii* seems to be significantly reduced at low temperatures when very few eggs are laid. This supports the results of other authors who found a complete absence of ovipositioning at temperatures of less than 12°C and that the maximum female feeding habit is at about 32°C (Parrella, 1984). However, at increased temperatures, the feeding habits and oviposition activity of females are significantly increased, with optimal behavior at about 30°C. These results confirm previous data showing that the optimal behavior of *L. trifolii* is at 24°C to 30°C (Heyer and Richter, 1990). On the other hand, that maximizes productivity and fecundity occurs at about 30°C (Miller and Isgar, 1985).

Naga *et al.* (2020) studied the relationship between *L. trifolii* and weather conditions and found that the number of mines, leaf infestation, and number of larvae were negatively correlated with minimum temperature (-0.61\*\*), evening relative humidity (-0.34\*), morning vapor pressure -0.55\*, evening vapor pressure -0.52\*, maximum temperature-0.57\*. Heyer and Richter (1990) concluded that the temperature sums and initial points of

development for the different stages of *L. trifolii* are 40.8; 14.0 (Egg), 31.5; 13.8 (Larva), 107.4; 13.6 (Pupa), and 172.5; 14.7 for the total development.

The relationship between *L. trifolii* population and both mean temperature and relative humidity was negatively insignificant in most growing seasons except night temperature which exposed high influence on *L. trifolii* population for larval and pupal stages recording (0.59 and 0.56) during the first season and (0.58 and 0.57) during the second growing season (Hady, 2004). Elkhouly (2009) concluded that a positive correlation between *D. isaea* numbers and mean temperature was estimated during five growing seasons, and a negative correlation were estimated between *D. isaea* numbers and mean RH. % in the winter seasons, while a positive correlations were estimated in the summer ones. A negative correlation was estimated between mean temperature and *O. pallipes* numbers during five growing seasons whereas positive correlations were estimated between *O. pallipes* numbers and mean RH. % during all growing seasons.

Every medicolegal death inquiry must include establishing the post-mortem interval (PMI). To carry out this estimation, a number of strategies based on various methodologies have been proposed. Among these, two of the methodologies evaluated the impact of the temperature and time on the parameters under consideration: accumulated degree days (ADDs), total body score (TBS), and insect development (Franceschetti *et al.*, 2021)

Therefore, it is very important to understand the population dynamics of *L. trifolii* and its essential biological antagonists *D. isaea* and *O. pallipes* under field conditions so that, the relationship between the population dynamics of *L. trifolii* and several climatic conditions should be studied. Furthermore, timely management

practices could be done and effective control measurements could be applied in greenhouse-controlled cultivations.

## Materials and methods

### 1. Population dynamics:

Surman region (Location: Latitude 32.7562 Longitude 12.5693) served as the site of the current investigation. In the growing seasons of 2020 and 2021, kidney bean (*Phaseolus vulgaris*) will serve as a summer host plant, and broad bean (*Vicia faba*) will serve as a winter host plant. From every host plant, 100 randomly selected leaflets were picked up each week. Samples were transferred to the laboratory for investigation while being maintained in plastic bags. We counted and recorded the number of infested leaflets, surviving *L. trifolii* larvae, and immature stages of the larval ectoparasitoid *D. isaea*. Under a stereo binocular microscope, infected leaves were examined in Petri dishes, and examined leaves were placed over moist filter paper. Daily or as needed, filter papers were wet to prevent leaflets from drying out until the appearance of *L. trifolii* or the larval pupal endoparasitoid *O. pallipes* was counted and recorded.

### 2. Metrological studies:

Daily records of mean temperatures along with relative humidity were obtained from the meteorological station of Tripoli to represent the climatic conditions effect, during 2020/2021 growing seasons. Mean values of temperatures and relative humidity were calculated, according to the following model:

$$Tm = \frac{T \max + T \min}{2}$$

Correlation coefficient values and linear regression equations were estimated using Microsoft Excel software 2016.

### 3. Degree day calculations:

Calculating degree days, *DD* was estimated through the use of daily maximum and minimum temperatures. The simplest method used to estimate the number of

degree days for one day is averaging. This averaging method ignores the upper threshold temperature: The basic equation for the calculation of degree days is established by McMaster and Wilhelm (1997)

$$DD = \frac{T \max + T \min}{2} - MMT$$

Where *DD* is the degree-days, *Tmax* and *Tmin* are the daily maximum and minimum temperatures, respectively, and *MTT* is the minimum temperature threshold estimated for *L. trifolii* by Lanzoni *et al.* (2002) and by Bazzocchi *et al.* (2003) for *D. isaea*.

## Results and discussion

### 1. Effect of mean temperature and relative humidity on the population of *Liriomyza trifolii*:

Data represented in Figures (1 and 2) and Table (1) showed positive correlations between the population of *L. trifolii* and mean temperature in the summer season and negative correlation in the winter season with *r* values (0.81 and -0.27). On the other hand, negative correlation values were calculated between the population of *L. trifolii* and relative humidity in the summer growing seasons (-0.29) and positive correlation in the winter season recording (0.109). The results are in line with those of Elkhoully (2003 and 2009). Furthermore, Minkenberg and Van Lenteren (1986) reported that, except for the temperature also the host plant had a great influence on the developmental time of *L. trifolii*. It was more rapid in kidney beans and slowed down in chrysanthemums and tomatoes by 3 and 6 days at a temperature of 20 °C. Heyer and Richter (1990) concluded that only in the very low (15°C) and in higher temperature ranges (33- 34°C) did the values of life cycle duration deviated more largely. These results are also in agreement with those of Hady (2004) who cleared that, the relationship between *L. trifolii* population and both mean temperature and relative humidity was negatively insignificant in most growing seasons.

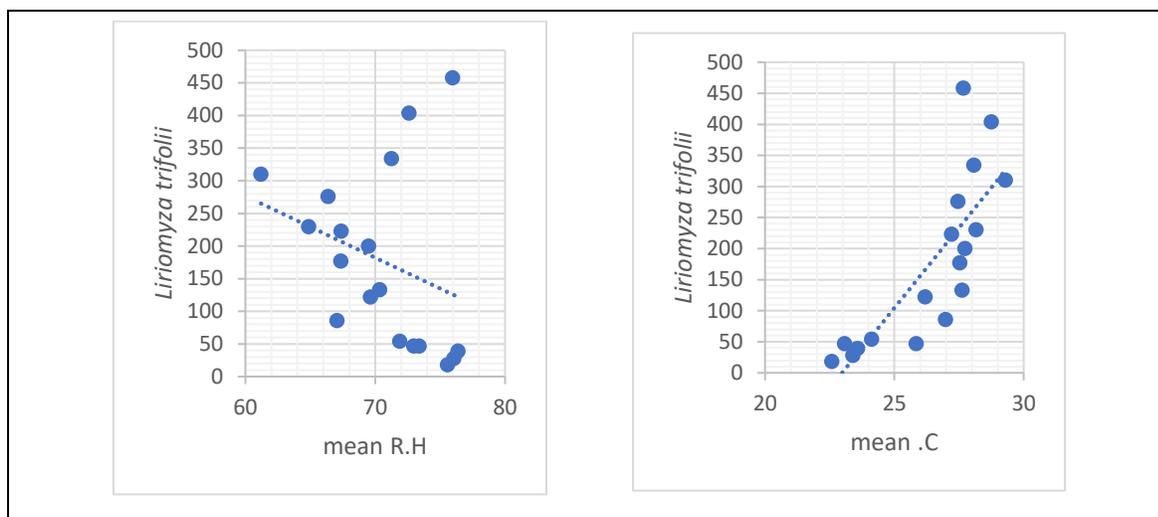


Figure (1): The liner regression showing the relation between mean temperature and relative humidity and population of *Liriomyza trifolii* on summer host plants.

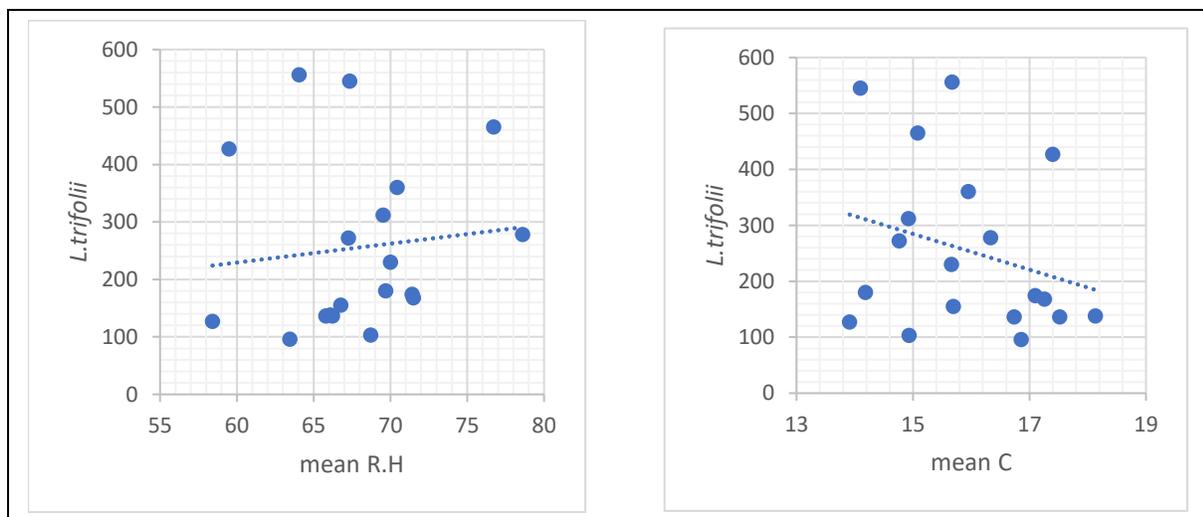


Figure (2): The liner regression showing the relation between mean temperature and relative humidity and population of *Liriomyza trifolii* on winter host plants.

Table (1): Linear regression equations and r values for the population of *Liriomyza trifolii* and temperature and relative humidity in summer and winter seasons.

Growing seasons	Mean temperature (C)		Mean R.H.	
	Regression equations	R	Regression equations	r
Summer	$y = 51.657x - 1187$	0.81	$y = -9.4473x + 843.43$	-0.29
Winter	$y = -31.94x + 763.74$	-0.270	$y = 3.3077x + 30.852$	0.109

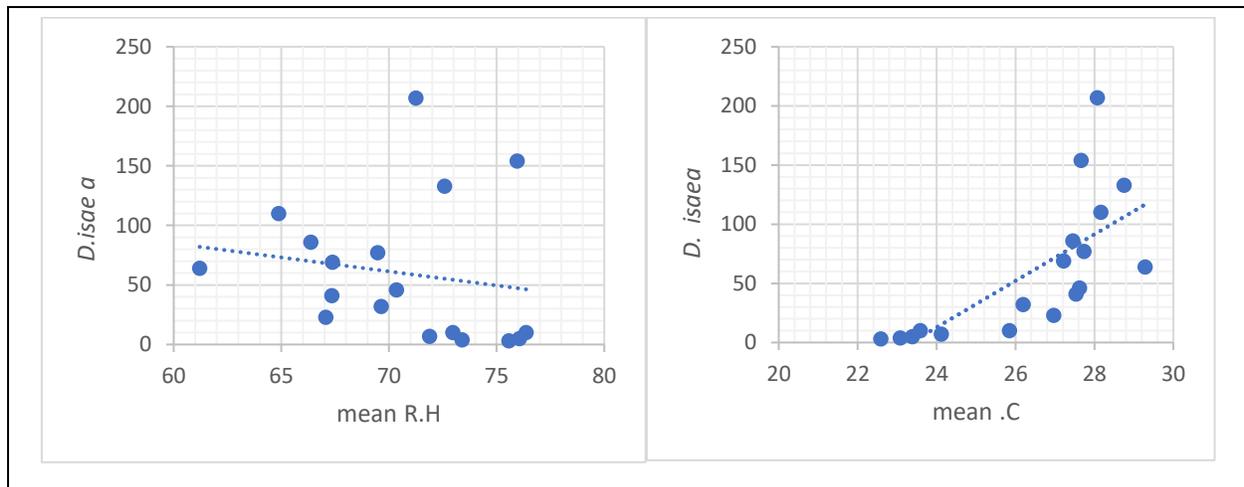
**2. Effect of mean temperature and relative humidity on the population of *Diglyphus isaea* :**

Data represented in Figures (3 and 4) and Table (2) showed positive correlations between the population of *D. isaea* and mean

temperature in the summer season and a negative correlation in the winter season with r values (0.70 and -0.1). On the other hand, negative correlation values were calculated between the population of *D.isaea* and relative humidity in the summer growing and positive correlations in the winter seasons with r values (-0.17, and 0.36), respectively, this finding could be explained by the thermal needs of *D.isaea* (Hondo *et al.*, 2006), who found that at the optimal range of development (15-25°C), the developmental period of *D.isaea* from egg to adult eclosion decreased as the temperature increased, so a positive correlation relationship was estimated between *D.isaea* populations and degrees of temperature, similar results were

obtained by Bazzocchi *et al.* (2003), so results in our study are in agreement with their proposal.

A low rate of RH. 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 *D.isaea* development in the summer growing seasons. It could be concluded that the positive correlation relationship that was estimated between mean temperature and *D.isaea* populations is due to the increase of developmental rate by the temperature increase in the results are in line with those of Elkhoully (2003, 2009) and Hady (2004).



**Figure (3): The linear regression showing the relation between mean temperature and relative humidity and the population of *Diglyphus isaea* on summer host plant.**

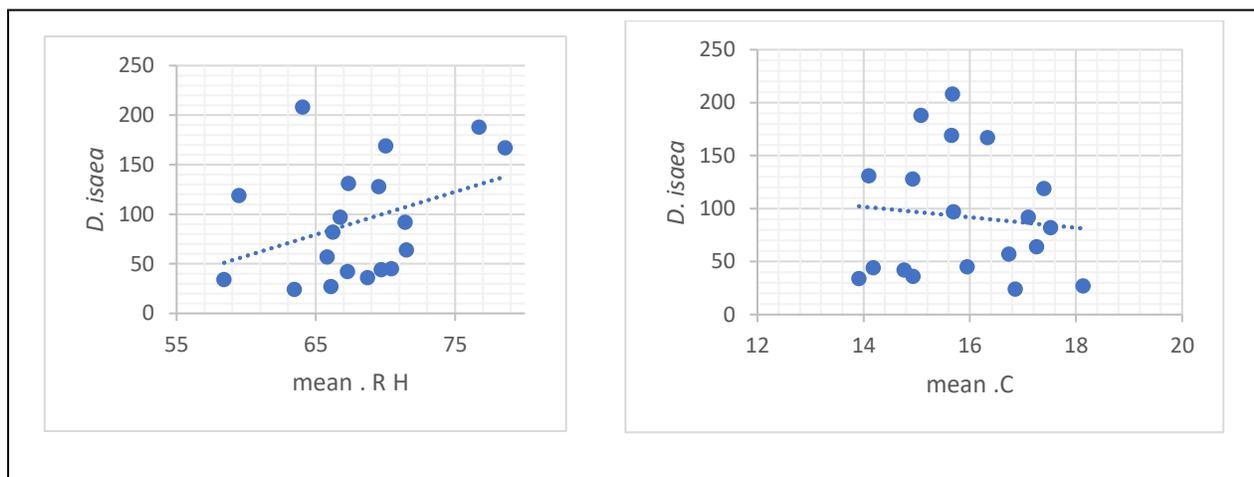


Figure (4): The linear regression showing the relation between mean temperature and relative humidity and the population of *Diglyphus isaea* on the winter host plant.

Table (2): Linear regression equations and r values for the population of *Diglyphus isaea* and temperature and relative humidity summer and winter growing seasons.

Growing seasons Seasons	Mean temperature (C)		Mean R.H.	
	Regression equations	R	Regression equations	R
Summer	$y = 19.661x - 459.08$	0.70	$y = -2.3528x + 226.02$	- 0.17
Winter	$y = -4.9689x + 171.35$	-0.10	$y = 4.3128x - 200.84$	0.36

### 3. Effect of mean temperature and relative humidity on the population of *Opius pallipes* :

Data represented in Figures (5 and 6) and Table (3) cleared a positive correlation between the population of *O.pallipes* and mean temperature in summer and winter seasons with r values (0.61 and 0.035). On the other hand, negative correlation values were calculated between the population of *O.pallipes* and relative humidity in the summer and winter seasons with r values (-0.30, and -.19), respectively.

It could be seen that a low temperature coincides with high relative humidity is a suitable climatic condition for *O.pallipes* development. *O.pallipes* occurred at high populations at the beginning of the winter growing seasons when mean temperatures were at their lowest values.

Ferro *et al.* (1979) demonstrated that an apple leaf under summer conditions could maintain its temperature at about 25°C over a range of an ambient air temperature of about 25-38°C through evaporate cooling. A similar results were estimated by Liebee (1984) who explained why the same development time occurred for the egg and the larval stages of *L.trifolii* at 30 and 25 in celery leaves because eggs and larvae of *L.trifolii* are inside the leaves. *O.pallipes* is an endoparasitoid so the increase of the ambient air temperature didn't affect the parasitoid populations so a negative correlation relationship could be estimated. Ferro *et al.*, 1979, Liebee, 1984 and Elkhouly, 2003 and 2009 could explain these results on cowpea leaves and broad beans in the summer and winter growing seasons when different temperatures have occurred.

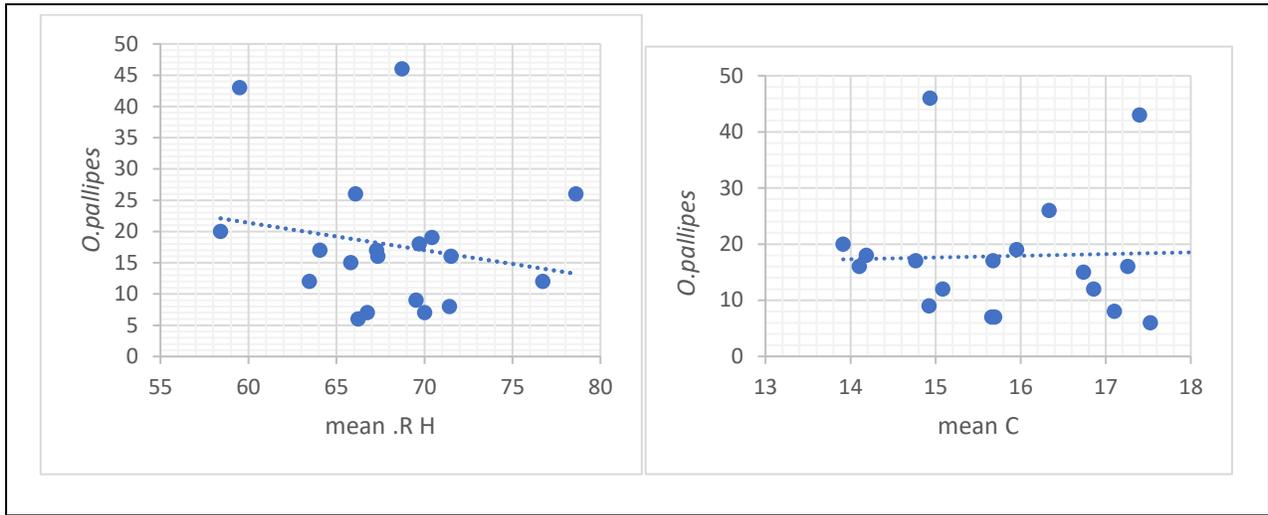


Figure (5): The linear regression showing the relation between mean temperature and relative humidity and the population of *Opius pallipes* on winter host plants.

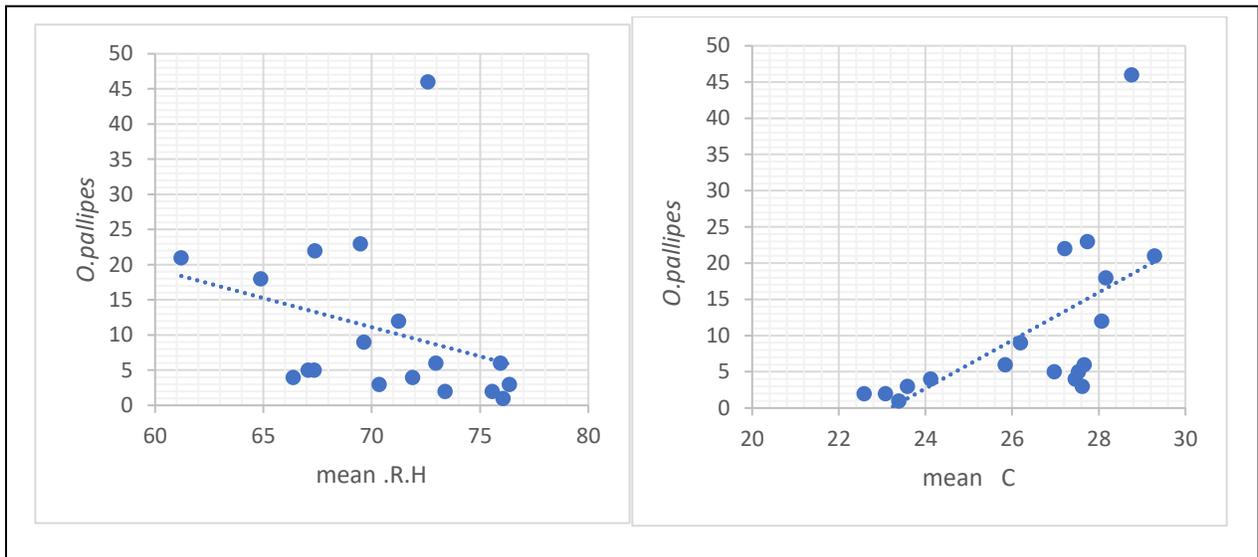


Figure (6): The linear regression showing the relation between mean temperature and relative humidity and the population of *Opius pallipes* on summer host plants.

Table ( 3): Linear regression equations and r values for the population of *Opius pallipes* and temperature and relative humidity during winter and summer seasons on broad bean (in winter) and kidney bean (in summer).

Growing seasons Seasons	Mean temperature (C)		Mean R.H.	
	Regression equations	r	Regression equations	r
Summer	$y = 3.3166x - 76.906$	0.61	$y = -0.8279x + 69.07$	-0.30
Winter	$y = 0.3059x + 13.029$	0.035	$y = -0.4389x + 47.729$	-0.19

#### 4. Population dynamics and degree days

##### DD calculation:

##### 4.1. Population dynamics of *Liriomyza trifolii*:

Data presented in Figure (7) clearly that, *L. trifolii* larvae recorded low numbers at the beginning of the season in early June, then the population increased recording three peaks of abundance (230, 458 and,

404 individuals/100 infested leaflets) on kidney bean during the summer growing season. On the other hand, *L. trifolii* larvae recorded low numbers at the beginning of the season in early December, then the population increased recording three peaks of abundance (168, 565, and, 312 individuals/ 100 infested leaflets) on the broad bean during the winter growing

season. Similar results were obtained by **Elkhouly et al. (2022)** they recorded three peaks of abundance for *L. trifolii* in summer and winter on two weed host plants, they also recorded a positive correlation value for *L. trifolii* and its associated parasitoids in both summer and winter growing seasons.

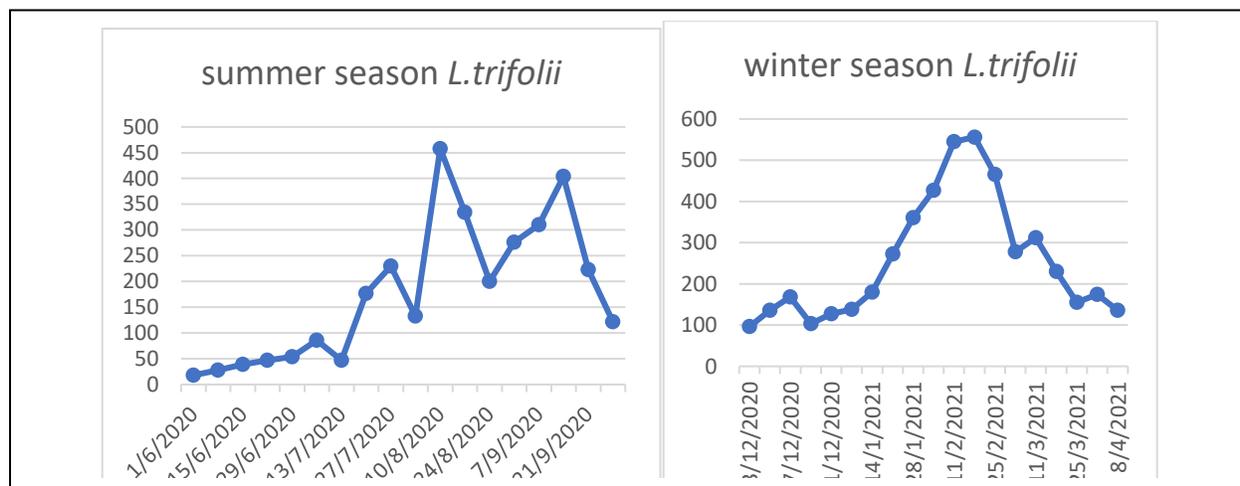


Figure (7): Population dynamics of *Liriomyza trifolii* on kidney bean and broad bean during summer and winter growing seasons 2020/2021.

**4.2. Population dynamics of *Diglyphus isaea* :**

Data presented in Figure (8) clearly that, *D. isaea* larvae were recorded in low numbers at the beginning of the season in early June, then the population increased in recording three peaks of abundance (110, 207, and, 133 individuals/ 100 infested leaflets) on kidney bean during the summer growing season. On the other hand, *D. isaea* larvae in recorded low numbers at the beginning of the season in early December, then the population increased recording three peaks of abundance (67, 208, and 169 individuals/ 100 infested leaflets) on the broad bean during the winter growing season. Elkhouly et al. (2015) concluded that the larval parasitoid *D. isaea* is the most dominant of the parasitoid complex of *L.bryonia*. The percentage of natural parasitism by *D. isaea* reached more than

50% in all studied host plants, so it seems to be a very effective natural antagonist on the tomato leaf miner *L. bryonia*, and could easily keep its populations at low levels ( Elkhouly et al., 2022).

The presence of parasitoids during all the periods of cultivation and the increase of this number in proportion to the increase in larvae. Similar to that observed for *L. trifolii* on beans the abundance of the parasitoids was positively correlated with leaf-miner host density (Li et al., 2012).

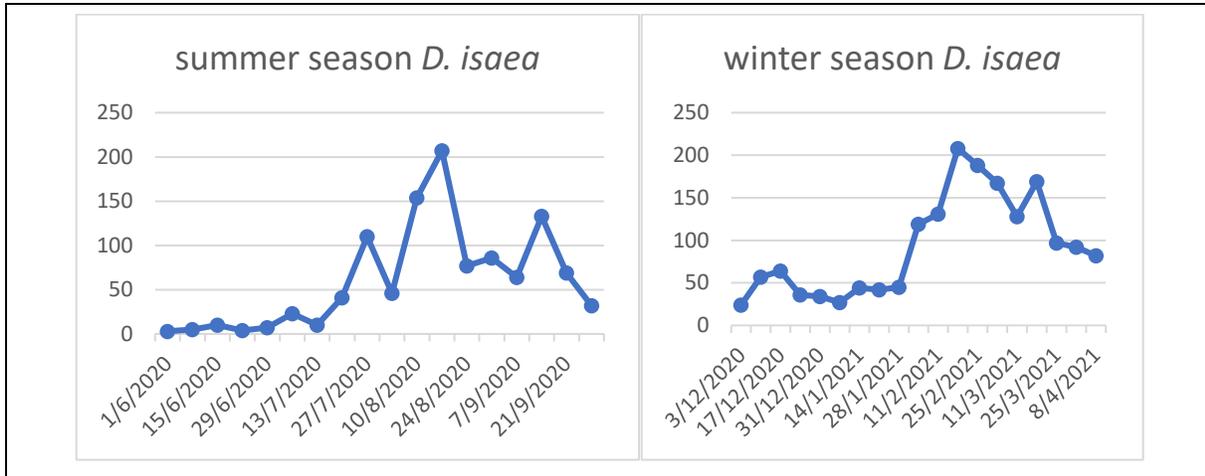


Figure (8): Population dynamics of *Diglyphus isaea* on kidney bean and broad bean during summer and winter growing seasons 2020/2021.

4.3. Population dynamics of *Opius pallipes* :

Data presented in Figure (9) clearly that, *O. pallipes* recorded in low numbers at the beginning of the season in early June, then the population increased recording four peaks of abundance (86, 230, and, 458 individuals/ 404 infested leaflets) on kidney bean during the summer growing season. On

the other hand, *O. pallipes* larvae were recorded in low numbers at the beginning of the season in early December, then the population increased recording three peaks of abundance (168, 565, and 312 individuals/ 100 infested leaflets) on the broad bean during the winter growing season.

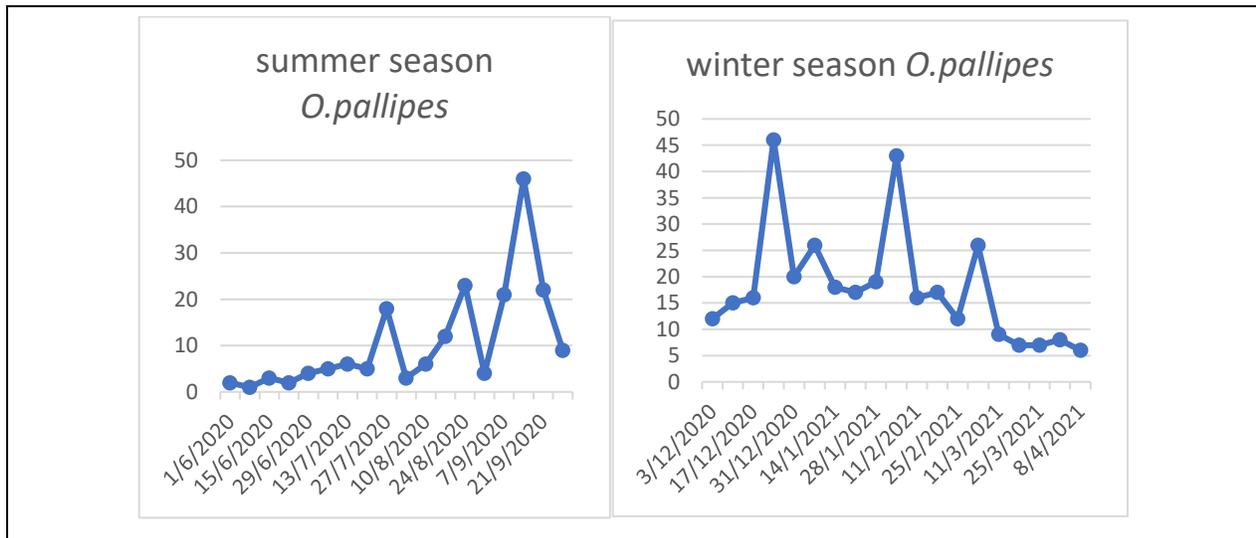


Figure (9): Population dynamics of *Opius pallipes* on kidney bean and broad bean during summer and winter growing seasons 2020/2021.

5. Degree day estimation of *Liriomyza trifilii* and *Diglyphus isaea*:

As presented in Table (4) the expected generations of *L. trifilii* and *D. isaea* by Lanzoni *et al.* (2002) and Bazzocchi *et al.*

(2003 ) estimated 2.999 and 5.51 generations for the winter season. These findings are in line with our results which estimated 3-4 peaks of abundance for *L. trifolii* and *D. isaea* for every successive season during the course of the study. On the other hand, a number of generations recorded for *D. isaea* in the

present study was lower than the estimated values suggested by the former authors, a possible explanation is the climatic circumstances under the Libyan conditions which may record a great variation of the air temperature and relative humidity during the growing season.

**Table (4): Degree day estimation of *Liriomyza trifolii* and *Diglyphus isaea* during the winter growing season.**

Insect	Accumulative.DD over the lower threshold	Estimated DD	Estimated lower threshold	Expected generations
<i>Liriomyza trifolii</i> Winter)	719.65	233.9 (Lanzoni <i>et al.</i> , 2002)	10.5 (Lanzoni <i>et al.</i> , 2002)	2.999
<i>Diglyphus isaea</i> (Winter)	891.96	161.8 (Bazzocchi <i>et al.</i> , 2003)	9.2 (Bazzocchi <i>et al.</i> , 2003)	5.51

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