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RESEARCH ARTICLE
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Epidemiological Prediction Study of Olive Fly *Bactrocera oleae* (Rossi)

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ABSTRACT

The olive (Olea europaea L.) is Jordan's most important fruit crop. Among the most serious insect pests is the olive fruit fly, *Bactrocera* (*Dacus*) *oleae* (Rossi). Two experiments were conducted to address this issue: a field experiment to determine the period of adult fly activity and its numerical fluctuations, and another experiment was performed in the laboratory to determine the duration of fly generation at two constant temperatures, as well as to calculate the critical limit for growth and the thermal constant. The fly's epidemiology was predicted during its activity period across different seasons using data from the experiments, average daily temperature data from the Meteorological Department, and observations of the phenological development of olive fruits. Results revealed two activity peaks: a summer peak in July and a fall peak in October, with the activity period lasting 242 days. The generation time was 28 days at 20±2°C and 18 days at 30±2°C. The pest grows at temperatures above 2°C and requires 504°C/day, with 9–10 generations per year. Therefore, it is crucial to monitor average temperatures in olive fields to predict the possible outbreaks of this pest and develop appropriate management practices.

Keywords: *Bactrocera oleae*, Critical growth limit, Jordan, Olive, Thermal constant.

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INTRODUCTION

The olive (*Olea europaea* L., family: Oleaceae) is the most important fruit crop in Jordan (International Olive Council (IOC) 2018). Jordan is among the globally top 10 olive-producing countries, with more than 20 million trees across the kingdom, covering 130,000 hectares of the country's total area (FAO, 2021; IOC, 2021; Freihat et al., 2021). The olive sector constitutes about 20% of the planted area in Jordan, which is one of the natural regions for olive cultivation in the Middle East. Olives cultivated in mountainous, plain, and desert areas of Jordan, with 60% in the northern region, 32% in the central region, and 8% in the southern region. Investments in the olive sector exceed one billion Jordanian dinars. The planted area has increased to 57000 hectares, and the number of trees has reached about 11 million, accounting for 72% of the fruit tree planted area and 20% of Jordan's total cultivated area (International Olive Council, (IOC) 2023). All of these made it possible for Jordan to become self-sufficient in olive production; approximately 102.4% of local demand (Department of Statistics of Jordan (DOS), 2021).

However, olive cultivation and production face many challenges, the most significant of which are pests. Among the most serious insect pests is the monophagous olive fruit fly, *Bactrocera (Dacus) oleae* (Rossi) (Diptera: Tephritidae) (Nardi et al., 2005; Al-Raddad & Mustafa, 2008; Burrack et al., 2011; Preu et al., 2020; Boulahia-kheder, 2021a,b). The female deposits eggs inside developing olive fruits; these eggs hatch two to three days later, and the larvae begin feeding on the olive flesh (Fletcher, 1987; Rice, 2000; Nardi et al., 2003). The larval stage lasts around 20 days (Rice, 2000). Pupation can occur within the olive fruit or in the soil, with pupation within the fruit lasting around 8–10 days (Vossen & Kicenik Devarenne, 2006; Heve et al., 2016). Unlike the larvae, adults do not feed on olives but on nutrient-rich substances such as honeydew and bird droppings

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(Rice, 2000). The female olive fruit fly can lay up to 500 eggs during its lifetime, which spans less than six months (Rice, 2000). In warm areas with abundant host plants, the olive fruit fly may produce up to five or six generations per year (Rice, 2000).

The oviposition activity of the female, as well as larval feeding, are responsible for the destruction of olive fruits. Additionally, the insect's oviposition behavior increases the fruit's susceptibility to bacterial and fungal pathogens (Delkash-Roudsari et al., 2014). One of the main challenges facing olive plantations worldwide, as well as in Jordan, is olive knot disease. *Pseudomonas savastanoi* pv. *savastanoi* (Smith 1908) caused the disease, and opinions on the olive fly's ability to spread it is varying. In North America, the disease spreads via pruning tools, infected seedlings, raindrops, and wind, with no insect vector involved, while in Europe, the olive fly plays a role in spreading the disease (Adaskaveg et al., 2012).

Infested olive fruits completely lose their market value for table consumption and oil production (Tzanakakis, 2003; Rice, 2000). Table olives have no tolerance for fly infestation, whereas oil olives tolerate a threshold of roughly 10% infestation, which can produce high-quality oil if processed quickly (Kicenik Devarenne & Vossen, 2007).

In order to effectively manage and control the olive fly, researchers studied its activity periods and numerical fluctuations in the field, conducted a laboratory study to determine its generation duration, calculated the critical limit for growth, and determined its thermal constant (Niassy et al., 2022).

MATERIALS & METHODS

Field Experiment

Two locations in Amman, Jordan, were chosen to conduct the field study because they were thought to have olive knot disease (*Pseudomonas savastanoi* pv. *savastanoi*): one was in the Village of Alnair (Latitude: 31° 56' 58" N, Longitude: 35° 49' 11" E) in Wadi Al-Sir area. The other location was on the campus of the University of Jordan (Latitude: 32° 0' 49" N, Longitude: 35° 52' 23" E) in Al-Jubeiha area (Fig. 1). Eight conical plastic traps were hung at each site. The lower half of the trap was yellow, designed as an inverted funnel containing a material that attracts the adult olive flies, while the upper half was transparent, acting as a cover for the funnel. The upper section also featured a fixed part that allowed for the suspension of the trap. Apak and Baspinar (2017) filled traps with 250 ml of an attractant solution made up of DAP fertilizer, amino phosphate, yeast, and water. The traps were randomly distributed on olive trees infected with olive knot disease, hanging each trap on one of the main branches at a height of 1.5–2 meters from the soil surface. The distance between traps was sufficient to prevent their attractive effects from interfering with each other (Apak & Baspinar, 2021).

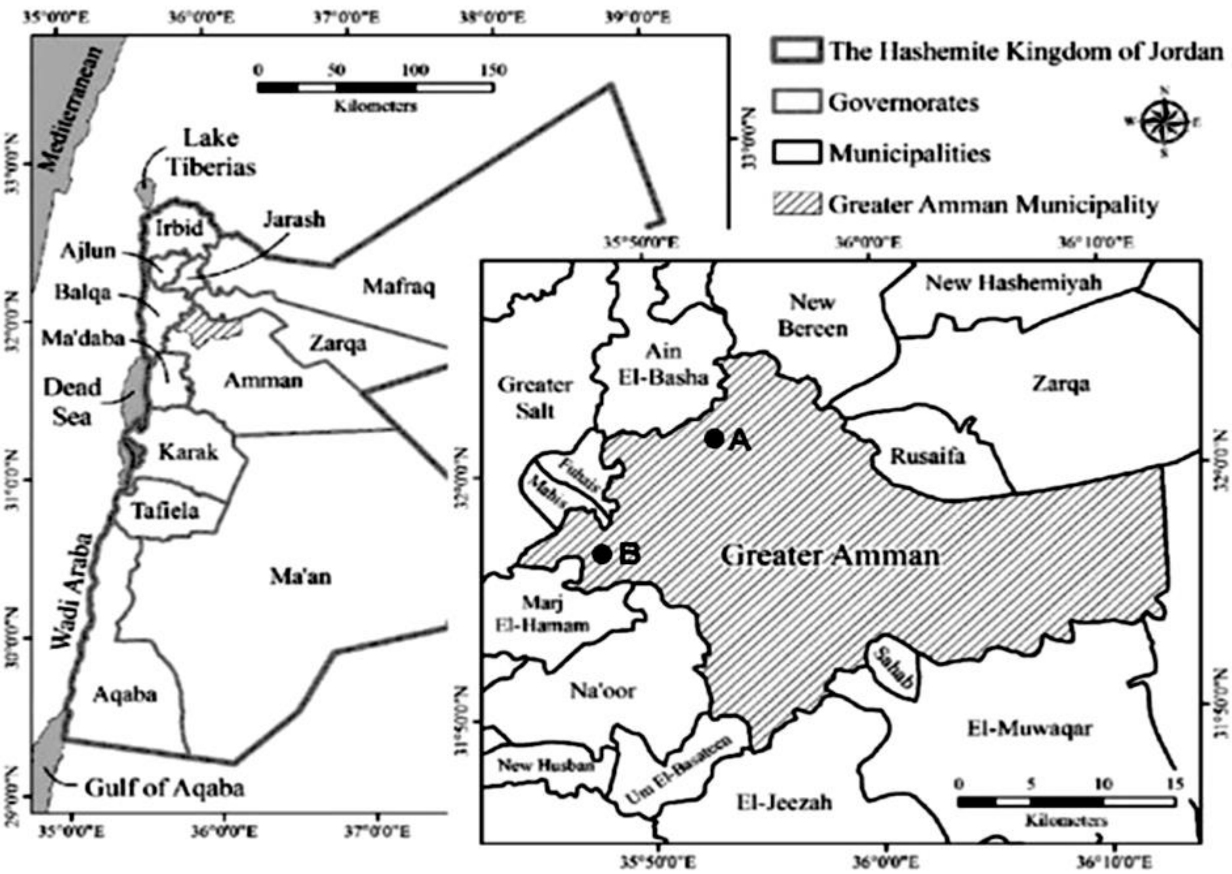

Fig. 1: Location map of Amman Governorate in the Hashemite Kingdom of Jordan. Solid circles represent sampling locations; (A) Al-Jubeiha (Latitude: 32° 0' 49" N, Longitude: 35° 52' 23" E), (B) Alnair Village (Latitude: 31° 56' 58" N, Longitude: 35° 49' 11" E) in Wadi Al-Sir area.

On February 7, 2022, the traps were hung at both locations. The contents of each trap were emptied into a filter after a week or two to prevent the adults from passing through. The adult insects were counted using a hand lens with a 20x magnification power. The traps then were cleaned with water, refilled with the attractant solution, and rehung on the same tree. This process was repeated at regular intervals from 1 to 2 weeks- depending on the number of insects caught- until the olives were harvested on October 17, 2022, in Alnair village and on December 28, 2022, on the University of Jordan campus. The number of flies caught in the eight traps at each site during each period was recorded (Table 1). The average daily temperature during each trapping period was calculated (Table 1). The time-related fluctuations in insect numbers were graphically illustrated in Fig. 2.

Table 1: Fluctuation in olive fruit fly population and the average daily temperatures from February 2, 2022, to November 19, 2022, in Alnair, Wadi Al-Sir area, and the University of Jordan campus, Jubaiha area

Sampling Date	Alnair village/ Wadi Al-Sir area		The University of Jordan campus/ Jubaiha area	
	number of flies/8 traps	Average daily temperature (°C)	number of flies/8 traps	Average daily temperature (°C)
February, 17	9	6.9	*	*
March, 2	9	9.9	*	*
March, 22	6	10.9	1	10.8
April, 11	9	13.3	0	13.3
April, 28	7	18.8	3	18.8
May, 16	4	21.6	4	21.6
June, 2	5	21.5	3	21.5
June, 14	8	23.7	5	23.7
June, 20	10	28.0	9	*
June, 24	11	*	38	26.1
June, 29	32	25.5	46	*
July, 5	19	27.2	74	25.1
July, 13	11	25.1	40	27.3
July, 17	9	*	24	27.0
July, 27	5	27.6	3	27.9
August, 8	4	*	8	28.9
August, 16	5	29.7	26	28.9
September, 5	7	26.5	12	29.2
September, 19	2	26.7	20	26.0
October, 17	10	34.4	43	25.9
October, 24	13	*	104	22.5
November, 10	5	*	201	25.0
November, 19	2	*	48	23.1

*No reading was taken.

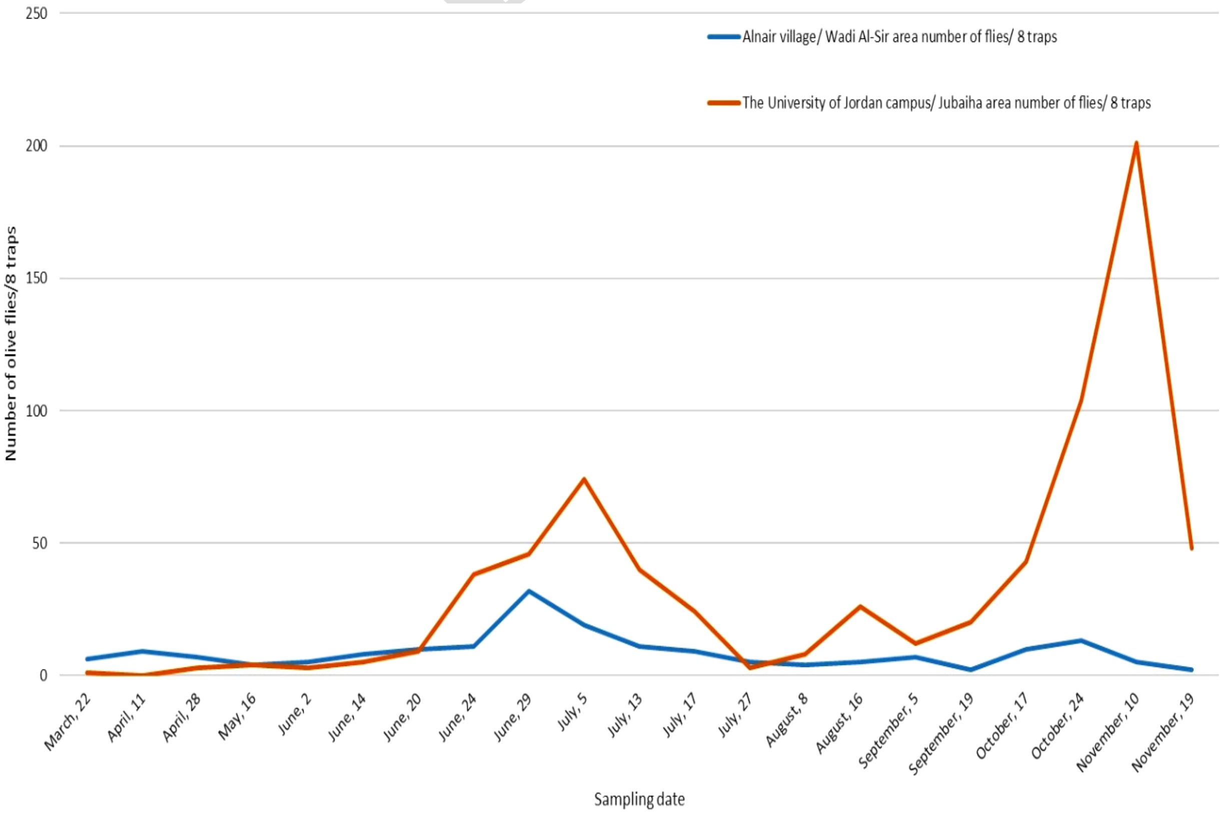

Fig. 2: Fluctuation in olive fly population during the period of olive growing season 2022 in Alnair in Wadi Al-Sir and campus of the University of Jordan/ Jubaiha area.

Laboratory Experiment

A large quantity of infested olive fruits was collected from both sites. The fruits were divided into two halves and placed in plastic containers with a 10 cm height edge. The containers were lined with shrunken papers to absorb moisture and create a suitable substrate for pupation. The containers were covered with a piece of voile cloth to ensure adequate ventilation, and sealed the lid with a straw. Strips of honey were placed on the lid's internal surface to provide food for the emerged adults. The first container was placed in an incubator at 20±2°C, while the second was placed in an incubator at 30±2°C. The containers were monitored until the adult insects emerged from the pupae and collected using a vacuum device. After placing the insects in refrigerators to reduce their activity, insects were sorted into males and females.

Twenty-five of each males and females were took, then placed in a plastic container with the same specifications as mentioned before, but this time it contained non-infested olive fruits. The container was placed in an incubator at 20±2°C. Similarly, another 50 males and females were placed in another container containing healthy olive fruits placed in an incubator at 30±2°C. A random sample of olive fruits was daily took and dissected using a scalpel and needle to check for eggs and larvae to track the growth and development of these stages. When larvae completed their development, they emerged from the olive fruits and started pupating under the shrunken paper at the bottom of the container. The adult emergence was observed on the underside of the container lid, feeding on the honey. The experiment was repeated three times at 20±2°C, which corresponds to the average temperatures in the spring and/or autumn seasons. It was also conducted once at 30±2°C, which corresponds to the average summer temperature. The time periods of growth and development for each stages were recorded at both temperatures. The periods required for the development of the different stages were summed and the generation duration was calculated (Table 2).

Table 2: Average growth periods of the immature stages of the olive fruit fly and the duration of a generation at two constant temperatures.

Temperature (°C)	Average growth period (day)			Generation duration (day)
	Eggs	Larvae	Pupae	
20± 2	6	14	8	28
30± 2	3	10	5	18

Using the equation: K = N (T – t), where:

- K is the thermal constant, measured in degree-days.
- N is the generation duration.
- T is the average daily temperature in degrees Celsius.
- t is the critical limit for growth (the temperature at which growth score is zero).

The critical limit for growth (t) and the thermal constant (K) were calculated, representing the effective temperatures required for the insect's growth and development. The value of K is constant regardless of temperature differences. We also calculated the average daily temperature for both seasons (Table 1). We determined the generation period and the number of generations per year accordingly.

Samples showing olive knot symptoms were collected. Disease causal agent was isolated and identified through biochemical LOPAT tests, followed by pathogenicity tests on olive seedlings of the K18 cultivar, as recommended by Schaad et al. (2001).

RESULTS

Olive fly activity in Wadi Al-Sir area/ Alnair village commenced on February 17, 2022- capturing of nine insects- and persisted until October 17, 2022, when fruits were harvested. This results in a total activity period of 242 days (11+31+30+31+31+30+17). In the Jubeiha area, on the University of Jordan campus, the insect began its activity on March 22, 2022, with only one insect caught. The activity continued until November 19, 2022, with olives harvested later on December 28, 2022. This also results in an activity period of 242 days (9+30+31+30+31+31+30+31+19). Thus, the pest's activity period is consistently 242 days at both sites.

It is also apparent that the insect population in both sites remained small until June 20, 2022 (Fig. 2), with the number of flies caught ranging between zero and nine insects per eight traps. After this date, the population began to increase at the University of Jordan site, reaching a summer peak of 75 flies on July 5, 2022. The population then decreased, peaking again in the fall with 104 flies on October 24 and 201 flies on November 11, 2022.

It is clear from Table 2 that the olive fly has completed its life cycle within one generation (the emergence of the adult insect until it re-emerges again) after 28 days at a temperature of 20±2°C and after 18 days at a temperature of 30±2°C, and by applying the equation: K= N (T – t):

Then K = 28 (20 - t) = 18 (30 - t).

= 560 – 28t = 540 – 28t

28t – 18t = 560 – 540

10t = 20

t = 20/10 = 2°C

Substituting the value of (t) into the equation and using any of the average daily temperatures:

K=28(20–2) = 504 day-degrees

or

K=18(30–2) = 504 day-degrees.

This means that the olive fly begins to grow at temperatures above 2°C and needs 504°C/day—i.e., a number of days during which the temperature exceeds 2°C, totaling 504°C/day—to complete one generation.

Given that the average daily temperature in the winter season in Alnair village/ Wadi Al-Sir area is 6.9°C, the duration of one generation during that season is:

504/6.9=73 days.

For the spring and autumn seasons at both locations, with an average daily temperature of about 18°C, the generation period is:

504/18=28 days.

In the summer, when temperatures rise and the daily average is about 26°C, the generation period will be:

504/26=21 days.

The number of fly generations per year was calculated by calculating the average daily temperatures at each site, determining the duration of a generation, and dividing the fly's activity period by the generation's duration.

In Alnair village/ Wadi Al-Sir area, the average daily temperature during the insect's activity period was 21.6°C, resulting in an average generation duration of:

$504 = N (21.6-2) = 19.6N$

$N = 504/19.6 = 25.7$ days N.

Thus, the number of generations will be:

$242/25.7 = 9.4$ generations.

At the University of Jordan campus/ Jubaiha area, the average daily temperature during the insect's activity period was 23.7°C, resulting in an average generation duration of:

$504 = N (23.7-2) = 21.7N$

$N = 504/21.7 = 23.2$ days

The number of generations is therefore:

$242/23.2 = 10.4$ generations

This indicates that the olive fly has 9–10 generations per year.

All olive trees used in this research, which were suspected to be infected with olive knot disease, were confirmed to be infected with *Pseudomonas savastanoi* pv. *savastanoi* based on biochemical and pathogenicity tests.

DISCUSSION

Precise ecological pest measurement parameters and calculations are essential for effective pest control. These parameters include the activity period, growth rate, and the number of generations under specific environmental temperatures. All this information was calculated after collecting data in the field and laboratory experiments. The study enables better predictions of when adults are active and when olives are at risk. Olive fly control methods typically rely on adult monitoring using various traps, such as yellow sticky traps, McPhail traps, etc., and assessing infested fruits to make informed decisions on fly control (Dias et al., 2018; Burrack et al., 2008). Decision-makers in pest control can benefit from the data from field traps (Pontikakos et al., 2012).

Field experiments in this study showed that olive fly activity began in mid-February to the last third of March and continued from mid-October to the end of the second trimester of November. The same is true in inland areas of California where almost the same climate prevails, adult flies emerge from March to May. As the new olive crop develops, females begin to lay eggs and become attracted to the fruits by late June to early July (Zalom et al., 2009; Yokoyama, 2015).

The current results revealed that the pest has two activity peaks: one in the summer (beginning of July) and another in the fall (end of October), with a total activity period of 242 days. These findings align with those of Burrack et al. (2011), where traps showed fly peaks in the summer and fall in California, spring peaks occur in March, April, or May, and fall peaks are in September, October, or November, while Apak and Baspinar (2021) reported only one fly peak in the fall. Ordano et al. (2015) proposed that a special reproductive mechanism, such as reproductive quiescence, allows populations of monophagous fruit flies like the olive fly to remain stable over time.

The two activity peaks of the olive fly were consistent with the highest epiphytic population dynamics of the olive knot bacterium, as reported by Wreikat and Khlaif (2017). They observed that the spring and fall seasons recovered the highest epiphytic olive knot bacterium population, with newly formed knots following insects peaks. Thus, it is believed that there is a relationship between the olive fly activity and the olive knot pathogen epidemiology.

The olive fly's generation time was 28 days at a temperature of 20±2˚C and 18 days at a temperature of 30±2˚C. The fly begins to grow at temperatures above 2˚C and requires 504°C/day—i.e., a certain number of days where the temperature exceeds 2˚C to complete one generation. Ordano et al., (2015) found that the temperature in olive groves was the main exogenous driver of the pest population. This finding is in agreement with Katsikogiannis et al., (2023), who reported that climate and altitude are the factors that most affect olive fly populations throughout the growing season. Mohammadipour et al. (2022) found that the heat requirements for the total immature stages of the olive fly were 348.51, 396.94, and 338.40 degree days in Siahpoush, Qushchi, and Kallaj, respectively, using the Ikemoto model. They also estimated the low temperature threshold for growth to be 9.31, 8.41, and 9.39°C for the total immature stages in the 3 fore-mentioned regions, respectively.

However, the current study's results do not match up with Mohammadipour et al. (2022) findings for the olive fly's temperature needs. This could be because the fly subpopulations in the two studies' regions are different. The three subpopulations of the olive fly are the Western Mediterranean, Central Mediterranean, and Eastern Mediterranean (Zygouridis et al., 2009). Mohammadipour et al. (2022) study conducted in Iran, therefore, may be similar to Eastern Mediterranean subpopulation of the olive fly. Meanwhile, the current results closely align with Zalom et al. (2009), who reported that multiple generations of the olive fly occur in California throughout the summer and fall. In the summer, flies can complete a generation in as little as 30 to 35 days, given optimum temperatures (20 to 30˚C) (Zalom et al., 2009). Furthermore, as shown in the current study, the pest begins to grow at temperatures above 2˚C. Zalom et al. (2009) also found that the lower and upper adult activity thresholds for the olive fly are about 15.6˚C and 17.2˚C, respectively, and below or above these thresholds, the flies are not active enough.

Our findings showed that winter fly generations took 73 days (based on the average daily winter temperature of 6.9°C), spring and autumn generations took 28 days (based on the average daily spring and autumn temperature of 18°C), and summer generations took 21 days (based on the average daily summer temperature of 26°C). The number of fly generations per year was 9.4 in the Wadi Al-Sir area and 10.4 in the Jubeiha area, indicating that the pest has 9–10 generations per year. However, the results of Burrack (2007) contradict the results of the current study in terms of the number of generations, as they found that the fly has at least four generations per year in California, with a partial generation spanning the winter.

Proposed Solutions to Manage the Olive Fly

Results showed that the fly emerges as a pupa during the winter and after fruits harvesting. If the temperature rises above 2°C, the fly begins its growth, becomes active, and starts flying after mid-February, remaining alive for 242 days. The activity of this fly ceases when the olive season concludes. However, their numbers remain low until mid-June. The insect produces between 9 and 10 generations per year, and the duration of each generation varies according to the prevailing temperature. Therefore, we propose the following solutions to manage the olive fly effectively:

1. Spread traps in olive groves to detect the adult fly and monitor its activity and population fluctuations.
2. Avoid using insecticides before mid-June because the fly does not infest olive fruits until they reach a size similar to that of a chickpea seed, which occurs around mid-June. Additionally, the insect population is small and did not cause significant damage before this date.
3. Use insecticides after mid-June, as follows:
   - Inventory and classify natural enemies and assess their efficiency in controlling the fly and reducing its numbers.
   - Choose specialized and safe insecticides, or those that are less harmful to natural enemies.
   - Evaluate the results of control measures before repeating them.
   - Continue control operations according to the specifications of the insecticide.
   - Cease chemical control for a safe period to minimize the impact of insecticides on olive oil and their harmful effects on humans.
4. Focus control efforts on the pest's adult stage since it is the weakest stage in its life cycle. Controlling the other stages is difficult due to the presence of eggs and larvae inside the fruits and the high resistance of pupae to insecticides.

Conclusion

The olive fly, *Bactrocera (Dacus) oleae* (Rossi), is considered one of the main insect pests of olives across olive plantation areas, significantly affecting both the quantity and quality of olive fruits and oil. In Jordan, this pest has 9–10 generations per year, each generation lasting between 18–28 days, depending on the average temperatures during the olive growing season. Additionally, there is a notable coincidence between the activity peaks of the olive fly and the highest epiphytic population dynamics of the olive knot bacterium.

Conflict of Interest: The authors declare that there is no conflict of interest.

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Author's Contribution: The corresponding author performed the research design, execution, data analysis, interpretation, and manuscript writing. Prof. Ihab Ghabeish and Prof. Hamed Khlaif followed up, analyzed, and evaluated the experiments throughout the research period. They also made valued contributions to the discussion, revision, editing, and proofreading of the final manuscript.

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