# Changes in Chemical and Physical Quality Attributes of Olives as Influenced by Chemical Control of the Olive Fruit Fly, Bactrocera oleae (Rossi) (Diptera, Tephritidae) in Jordan

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### ABSTRACT

The olive fruit fly, Bactrocera oleae (Rossi) (Diptera, Tephritidae) is the most devastating insect pest possessing severe economic threats for the olive growers in Jordan. Therefore, the current study aimed at determining the effect of insecticidal applications against B. oleae on olive quantity, as well as physical (refractive index, fruit weight, diameter and volume, and weight of seed and flesh, as well as oil percentage) and chemical (olive oil acidity and peroxide value parameters). The experiment was conducted using Nabali olive cultivar in Karak District-Jordan in the year 2016. Three insecticidal spraying significantly (P<0.05) reduced olive oil acidity, peroxide value, and refractive index, and increased olive fruit's weight, diameter and volume, and seed, and flesh weight compared to the untreated trees. In conclusion, the application of insecticides against B. oleae improved the olive quantity and tested quality parameters.

Keywords: Olive fruit fly, olive, oil quantity, chemical and physical quality, chemical applications, Jordan.

#### **INTRODUCTION**

Jordan is focusing on olive trees' cultivation, and olive become one of the most important fruit trees (Shdiefat *et al.*, 2009), reflected by increased cultivated areas, in which olive trees cover about 74% of the total area planted with fruit trees in Jordan. In 2016, the Jordanian Ministry of Agriculture figures indicated that olive trees occupy 56,600 ha, and produce 125,150 tons of olive oil (Jordan Statistical Yearbook, 2019). Jordan ranks 10th worldwide from the

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standpoint of olive oil production. Some of the produced olive oil is locally consumed, and the rest is exported to the Arab Gulf, European countries, the USA, and other countries (Al-Saed *et al.*, 2010).

The olive fruit fly, Bactrocera oleae (Rossi) (Dip., Tephritidae) is considered the most devastating insect pest possessing severe economic threats for the olive growers in the Mediterranean basin (Zalom *et al.*, 2009; Daane and Johnson, 2010; Al-Zyoud, 2014a, b, 2015, 2017). B. oleae affects olive tree cultivation, causing serious negative qualitative and quantitative consequences with economic impacts and losses (Vossen and Devarenne, 2006, Skouras et al., 2007). The losses ( $\sim$ 30%) are due to premature drop of the infested fruits, direct larval pulp destruction, and an overall reduction in the olive oil quality (Pereira et al., 2004; Nardi et al., 2006; Zalom et al., 2009). If control measures, especially insecticides, are not taken, up to 40% of olive production might be lost (Pereira et al., 2004; Skouras et al., 2007). The oil obtained from olive fruits infested with B. oleae has 50-60% higher acidity than the un-infested fruits (Parlati et al., 1990). Furthermore, exit holes made by the pest larvae promote bacterial and fungal infections (Pereira et al., 2004). Furthermore, B. oleae-infested olive fruits have low oil phenolic concentration (Pereira et al., 2004; Tamendijari et al., 2004), leading to compromised stability of olive oil.

Pesticides helped the world meets growing food demand by increasing agricultural productivity through controlling agricultural pests. Worldwide, chemical insecticides made it possible to protect olives efficiently from B. oleae infestation (Haniotakis, 2005). The predominant method of B. oleae control has been the use of conventional insecticides (Al-Zyoud, 2014a, b, 2015). Insecticides have been effectively used over the last many years and ensured yields and benefits to the olive growers. However, protein hydrolyzate mixed with insecticides bait sprays has been used for many years against B. oleae (Manousis and Moore, 1987; Roessler, 1989). Furthermore, the effective management of B. oleae depends, in addition to insecticides, on the trapping of adults, harvest timing, fruit sanitation after harvest, and biological control (Wood, 2009).

The complete dependency, extensive and massive use of pesticides in modern agriculture caused their widespread diffusion to all environmental compartments including a wide range of organisms up to the humans (Samiee et al., 2009; Abang *et al.*, 2013. On the contrary, if pesticides are used correctly, and despite their disadvantages, they will remain an important component of integrated pest management. Therefore, the current study aimed at

determining the effect of insecticidal applications against B. oleae infestation on olive quantity as well as physical and chemical quality parameters (i.e., olive oil acidity, peroxide value and refractive index, fruits' weight, diameter and volume, and weight of seed and flesh, as well as oil percentage).

## MATERIALS AND METHODS Experimental location, conditions, and design

The experiment was conducted at a private olive orchard of 13-year-old own-rooted "Nabali" olive in Al-Jadaa, Karak District of Jordan (Latitude of  $31^{\circ}11$ " and Longitude of  $35^{\circ}42$ ") in the 2016 growing season. The experimental site is characterized by semi-arid conditions with a long-term annual average rainfall of 300 mm and is located at 750 m above sea level. The trees spaced 5x5 m, trained in a modified central leader training method, and received routine horticultural care. The trees were irrigated with potable water using a drip irrigation system with 1 m/plant, each delivering 4 L h<sup>-1</sup>.

Three insecticides used; Lambda-cyhalothrin (Karate®, Pyrethroid); Chlorpyrifos (Dursban®, Organophosphorus) and Fytoclean, Russell IPM (as a non-synthetic insecticide; A.I.: Potassium salt and fatty acids from plant oils (Hassan and AlZaidi, 2008). The insecticides are used at the recommended rate of application by their manufacturers. Each treatment (insecticide) comprised four olive trees and was replicated three times, and the experiment was set up in a Randomized Complete Block Design (RCBD). Tap water is used as a control treatment. To prevent spray drift of insecticidal applications, the treated trees were at least 10 m far from each other. Trees sprayed twice using a knapsack sprayer. The first application was carried out on July 1st, 2016, and the second one on August 1st, 2016. One Kg of olive fruits/tree was picked up from five different directions of each tree (Top, east, west, north, and south) from each treatment on October 15th, 2016. The effect of insecticidal applications on olive oil acidity, peroxide value and refractive index, fruits' weight, diameter and volume, and

weight of seed and flesh, as well as oil percentage, was measured as mentioned below.

# Oil extraction and determination of olive oil acidity, peroxide value, and refractive index

The process of extracting the oil present in olive flesh was carried out in a mill olive fruit and pressed by blender, and then damped in hexane at  $35-65^{\circ}$ C for 24 h overnight. Hereafter, the oil is separated by a Vacurotary evaporator, and oil percentage is calculated. To gauge oil acidity, 5 g of extracted olive oil were taken from each treatment (Karate, Dursban, Fytoclean, and control), and then 50 ml of a dissolving solution consisting of Ethanol: Diethylether at a ratio of 1: 1 (V/V) was added to the oil. The mixture was shacked using a shaker for 5 minutes and 5 ml of Phenolphthalein reagent solution was added. The blend was titrated using a digital titration by adding 0.1N of NaOH until the color turned pinkish, at this stage; the titration stopped. The following formula is used to estimate olive oil acidity:

# Acidity (%) = <u>Volume of NaOH×0.1×F</u> Weight of oil (g)

Where F equals 28.2 g/mol as oleic acid

To measure peroxide value, 5 g of oil is kept in an Erlenmeyer flask. Fifty ml of glacial acetic acid and chloroform mixture at a ratio of 3: 2 (V/V) were added, and the flask was thoroughly shaken. After that, 0.5 ml of potassium iodide solution was added, and the flask shook sing a shaker for 5 minutes again. Fifty ml of distilled water was added to the flask and the sample was titrated with sodium thiosulfate solution (0.01N) until the color disappeared using starch as an indicator (AOACI, 1995). The peroxide value measured according to the formula:

Peroxide value (meq O<sub>2</sub>/kg oil) =  $(V_1-V_2) \times N \times 1000$ W

Where V<sub>1</sub>: Sample titration in ml, V<sub>2</sub>: Blank titration in ml, N: Normality of sodium thiosulfate, and W: Weight of oil sample (g).

The oil refractive index is measured by the ABBE refractometer. The oil is taken and placed onto the device, and the refractive index of olive oil is read through the microscope.

Determination of fruit' weight, diameter, volume, seed, and flesh weight

A sample of 200 olive fruits was taken randomly and the average weights of fruit, seed, and flesh were measured via an electronic digital balance. The average fruit volume was measured as follows: Twenty olive fruits were taken randomly and placed in a cylinder with a known amount of water. The amount of increase in the water volume after adding the fruits into the cylinder was recorded. The average fruit diameter was measured by a Vernier Caliper.

#### Statistical analyses

To affirm the basic assumptions of the data to be analyzed, they were firstly tested for the normal distribution and the homogeneity of variance using the Barlett-test (Kohler *et al.*, 2002). The statistical analysis was performed using the MSTATC program. Data were analyzed using a one-way analysis of variance (ANOVA) (Zar, 1999). When significant differences are detected, means are separated using the Least Significant Differences (LSD) test at 0.05 probability level (Abacus Concepts, 1991).

# **RESULTS** Olive oil acidity, peroxide value, and refractive index

The effect of insecticidal applications on olive oil acidity, peroxide value, and refractive index is shown in figure 1. Insecticidal application significantly (P<0.05) reduced olive oil acidity (0.28-0.30%) compared to the control (0.35%) (Figure 1A). All insecticides significantly reduced oil peroxide value, in which application of Fytoclean gave the lowest peroxide value (2.7 meq O<sub>2</sub>/kg of oil) (Figure 1B). The obtained results in figure 1C revealed a significant decrease in refractive index in response to insecticidal treatment. A higher refractive index was obtained in untreated trees (1.46) than the treated trees with all insecticides tested (Karate, Fytoclean, and Dursban). It is worthy to mention that

there were no significant differences in the olive oil acidity, peroxide value, and refractive index among the three different insecticides tested.

#### Fruit weight, diameter, and volume

The application of insecticides (Karate, Fytoclean, and Dursban) significantly increased olive fruit weight compared to the untreated trees (Figure 2A). Olive fruit diameter is significantly affected by insecticides' application (Figure 2B). The application of insecticides significantly influenced Fruit diameter. Particularly when Karate (15.6 mm) was used. Results of olive fruit volume (Figure 2C) showed a significant effect of insecticidal treatment on fruit volume. Application of Karate and Fytoclean significantly increased fruit volume compared to other treatments.



Figure 1. Oil acidity percentage (A), peroxide value (B), and refractive index (C) of Nabali olive cultivar treated by different insecticides. *Different small letters* above bars indicated significant differences among the different treatments within the same tested parameter at *p*<0.05 (one-factor analysis of variance).



Figure 2. Fruit weight (A), diameter (B), and volume (C) of "Nabali" olive as influenced by different insecticides. *Different small letters* above bars indicated significant differences among the different treatments within the same tested parameter at p<0.05.

#### Seed and flesh weights and oil percentage

There was a significant effect of the insecticidal application on olive seed weight (Figure 3A). Application of all insecticides significantly increased seed weight (0.70-0.74 g) compared to the untreated trees (0.61 g). The statistical analysis of the obtained data indicated a significant effect of the insecticidal application on flesh

weight (Figure 3B). The flesh weight was significantly increased by application of the three insecticides (2.51-2.55 g) compared to the control treatment (2.17 g). Application of insecticides did not significantly affect the olive oil content (Figure 3C), and it was found that the highest numerically olive oil content in the Dursban treatment (18.3%).



Figure 3. Weight of seeds (A), flesh (B), and oil percentage (based fresh weight) (C) of "Nabali" olive as influenced by different insecticides. *Different small letters* above bars indicated significant differences among the different treatments within the same tested parameter at p<0.05

#### DISCUSSION

Olive fruits are seriously deteriorated by damage caused by B. oleae attack, which strongly alters the quantity and quality of olive fruits and oil (Wang et al., 2009; Mraicha et al., 2010; Medjkouh et al., 2016). In addition, the action of B. oleae attack is the main external agent responsible for unwanted metabolic processes in olives that lead to a subsequent reduction in olive quantity and oil quality (Kiritsakis, 1998). However, the acidity of olive oil is considered a basic character of the quality. In the present study, insecticidal application significantly reduced olive oil acidity compared to the untreated trees. In addition, the decreased oil acidity in the current study might be attributed to the decreased action of lipolytic activity of microorganisms that enter the holes made by B. oleae (Pereira et al., 2004). Gomez-Caravaca et al. (2008) reported an increased oil acidity increased as the degree of B. oleae damage increased, which is in agreement with the findings of the present study.

The peroxide value is an indicator to reveal enzymatic and oxidative deterioration in olive oil (Barone et al., 1994). Peroxide value, a measure of the primary oxidation products, increases in oils obtained from damaged olive fruits (Pereira et al., 2004). The current results indicated that all insecticides significantly reduced oil peroxide value, in which application of Fytoclean gave the lowest peroxide value. In this regard, Bubola et al. (2014) reported that the increase of olive oil oxidation is probably due to exposure to oxygen and the damage to fruit skin caused by B. oleae attack. The fruits became more susceptible to the effect of atmospheric conditions and, therefore to oxidation. The peroxide values obtained in the current investigation are in agreement with those obtained by Pereira et al. (2004) and Gomez-Caravaca et al. (2008) who found that peroxide value in olive significantly increased with increasing olive infestation by B. oleae. Olive oil extracted from B. oleae infested

olive fruits gave higher peroxide value due to the compounds formed during primary oxidation, mainly hydroperoxides when compared with olive oils extracted from healthy fruits (Laguerre *et al.*, 2007).

The current results showed a significant decrease in refractive index in response to insecticidal treatments compared to untreated trees (1.46). Refractive index is the measure of the thickness as well as purity or clarity of the oil (Ogungbenle and Afolayan, 2015). However, the refractive index values of olive oil depend on their molecular weight, fatty acids chain length, degree of unsaturation, and degree of conjugation. The refractive index value for different olive oils generally varies between 1.447 and 1.482 (Roodak et al., 2016). According to Alimentarius (2001), the refractive index of virgin olive oils ranges from 1.4677 to 1.4705. Refractive index value depends on the degradation and percentage of polar compounds formed during oxidation and hydrolytic reactions (Benedito et al., 2007). In general, the current results were in the same trend as those reported by Ozkan et al. (2008), in which the refractive index for Ayvalik, Domat, and Gemlik Turkish olive cultivars were 1.4688, 1.4678, and 1.4689, respectively.

The present findings illustrated that application of insecticides significantly increased olive fruit's weight, diameter, and volume as well as seed and flesh weights, and this might be because insecticides application decreased *B. oleae* infestation, which resulted in increasing all parameters tested. The current results are in agreement with the findings of Medjkouh *et al.* (2016), who found that fruit weight is negatively influenced by *B. oleae* attack. The latter authors observed that fruit weight decreased from 1.4 to 1.0g and 1.7 to 1.2g with the increasing infestation for the "Limli" and "Roulette" olives, respectively. The exit holes, made by the *B. oleae* favor the entrance and development of bacteria and fungi, which increase olive pulp damage (Tamendijari *et al.*,

2004), and this resulted in a reduction in fruit and flesh weights. Efe *et al.* (2009) reported fruit and seed weights of Memecik olive of 4.78 and 0.56 g, respectively. Ismail *et al.* (2010) found olive seed weights of 0.55, 0.59, and 1.1 g for Nabali Muhassan, Nabali Baladi, and Shami olive fruits, respectively. However, the differences among the current and previous studies might be due to olive cultivar, soil characteristics, irrigation, fertilization, environmental conditions, *B. oleae* infestation level, and whether insecticides were applied or not.

In conclusion, the application of insecticides against *B. oleae* has many positive features on both olive quantity and quality parameters tested. A management strategy against *B. oleae* should be considered to preserve the quantity and quality of the Jordanian olive table and oil for their labeling and marketing in the local and international market.

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# التغيرات في خصائص الجودة الكيميائية والفيزيائية للزيتون من خلال المكافحة الكيميائية لذبابة ثمار الزيتون في الأردن Bactrocera oleae (Rossi) (Diptera, Tephritidae)

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# ملخص

تعدُّ ذبابة ثمار الزيتون Bactrocera oleae (Rossi) من أكثر الآفات الحشرية التي تحدث ضرراً اقتصادياً كبيراً لمزارعي الزيتون في الأردن، ولهذا هدفت الدراسة الحالية إلى دراسة تأثير رش المبيدات الحشرية ضد ذبابة ثمار الزيتون على الإنتاج وعلى بعض الخصائص الفيزيائية: (معامل الانكسار، وزن وقطر وحجم الثمار، وزن البذور واللب، ونسبة الزيت) والكيميائية: (حموضة زيت الزيتون وقيمة البيروكسيد). وأجريت التجربة باستخدام صنف الزيتون نبالي في محافظة الكرك-الأردن في عام 2016. وتم استخدام ثلاثة مبيدات حشرية: وهي مبيد الكاراتيه، والفايتوكلين، والدورسبان في عمليات الرش، وتم رش أشجار الزيتون مرتين، وأدى رش المبيدات الحشرية إلى خفض حموضة زيت الزيتون، وقيمة البيروكسيد ومعامل الانكسار بشكل معنوي، وزيادة معنوية في وزن وقطر وحجم ثمار الزيتون البذور واللب مقارنة بالأشجار غير المعاملة بالمبيدات. وفي الختام، أدى استخدام المبيدات الحشرية ضد ذبابة ثمار الزيتون إلى تصون البذور واللب مقارنة بالأشجار

الكلمات الدالة: نبابة ثمار الزيتون، الزيتون، كمية الزيت، الجودة الكيميائية والفيزيائية، المكافحة الكيميائية، الأردن.

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