

Phytochemical Components, Antioxidant Properties, Antimicrobial Effects, and Bio-Aphicidal Prospects Against the Black Bean Aphid (*Aphis fabae* Scop.) of *Olea europaea* L. Leaves Extracts from Morocco

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ABSTRACT

The escalating phenomenon of bacterial resistance to antibiotics over time, coupled with the consequential detrimental effects of oxidative stress on cellular aging, and the usage of pesticides with varying degrees of toxicity, thereby impacting both human health and the environment, represents a pressing global concern. Consequently, researchers are compelled to identify novel biomolecules derived from plants and their derivatives that possess antibacterial, antioxidant, and insecticidal properties. The olive tree (*Olea europaea* L.), a fruit-bearing tree within the Oleaceae family, characterized by its olives, has been cultivated for millennia, particularly in Mediterranean regions, with its leaves being primarily employed for their multifarious therapeutic attributes. In this investigation, extracts were procured from olive leaves through employment of the Soxhlet apparatus, followed by the quantification of total polyphenols and flavonoids. The assessment of the antioxidant potential of these extracts was conducted using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. The antibacterial efficacy was evaluated via the disk diffusion method against six pathogenic bacterial strains, namely *Staphylococcus aureus*, *Escherichia coli*, *Acinetobacter baumannii*, *Salmonella* sp., *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*. Additionally, the *in vitro* aphicide activity of *Olea europaea* L. extracts were investigated at concentrations of 0.5%, 1%, 1.5%, 2%, and 2.5% against the black bean aphid, *Aphis fabae* Scop. The findings of this study suggest that olive leaf extracts exhibit robust antioxidant properties and display modest antibacterial activity against pathogenic agents. Hence, these extracts are strongly endorsed for their potential role as eco-friendly antioxidants, and owing to their modest yet efficient insect-repelling attributes, they can be utilized as a sustainable, low-impact insecticide in the ecological engineering approach to controlling black aphids in bean crops. Therefore, the utilization of olive tree-derived extracts is encouraged.

Keywords: *Olea europaea* L., extracts, antibacterial effect, antioxidant activity, insecticidal activity, *Aphis fabae* Scop., aphicidal.

INTRODUCTION

Oxidative stress is a well-documented contributor to cellular aging, with its associated damages

implicated in the development of serious pathologies, including cancer. However, the use of synthetic antioxidant molecules has come under scrutiny due to potential health risks (Sindhi et al., 2013).

As a result, there is a growing body of research focused on natural antioxidants, particularly polyphenols and vitamins derived from plant sources. This research often centers around the characterization of endemic plants, which have demonstrated diverse biological activities, with polyphenols being particularly noteworthy (Haddou et al., 2023; Jaber et al., 2018b; Taibi et al., 2023b).

Simultaneously, the increasing bacterial resistance to antibiotics presents a substantial global challenge (Haddou et al., 2023; Jaber et al., 2018b; Taibi et al., 2023b). In response, there is a pressing need for the identification of new biomolecules with potent antibacterial properties (Kandsi et al., 2022a). The use of pesticides, while designed to combat harmful organisms, poses a significant threat to human health. Disturbingly, a considerable proportion of these chemicals are recognized as carcinogenic, further exacerbating their negative impact on human well-being. This issue extends to the environment, as these pesticides can also harm the nervous and endocrine systems. Seeking alternatives, researchers are turning to plant-derived substances, which tend to degrade more rapidly in the environment, thereby reducing their toxicity to humans, animals, and minimizing their environmental footprint (Bagheri and Rahimi, 2014).

Morocco, with its rich and diverse flora, has a thriving medicinal plant sector (Bencheikh et al., 2021; Radi et al., 2023). The country's unique plant species are a valuable source of secondary metabolites with diverse chemical structures and a wide array of biological activities. Investigating and characterizing these activities remains an engaging and worthwhile pursuit for numerous studies (Kandsi et al., 2022b, 2022a; Taibi et al., 2023a). The olive tree, scientifically known as *Olea europaea* L., holds a special place in Mediterranean cultures. Belonging to the Oleaceae family, it is not only a source of olives but also serves as an aromatic and medicinal plant. Throughout history, its leaves have been utilized for various purposes, including wound disinfection, and they are esteemed for their rich content of phenolic compounds, which have well-documented health benefits (Addab et al., 2020; Al-Attar and Alsalmi, 2019). This study aims to delve into the active constituents found in ethanolic and methanolic extracts of Moroccan olive leaves. It seeks to comprehensively evaluate their antioxidant, antibacterial, and aphicidal activities, further contributing to our understanding of the potential benefits of these extracts.

MATERIALS AND METHODS

Plant material

The Picholine olive tree (*Olea europaea* L.) leaves, harvested in November from Morocco's Oriental region, underwent a meticulous process. Firstly, the leaves were carefully washed to eliminate any dust or impurities. Subsequently, they were dried at room temperature, in conditions devoid of light and humidity. Once thoroughly dried, an electric mill was employed to grind the leaves into a vibrant green powder. The resulting product was then stored in hermetically sealed glass containers at a temperature of +4°C, ensuring protection against both light and humidity until the time of utilization.

Preparation of extracts

To obtain extracts from olive tree leaves, we employed the Soxhlet extraction method. The selected solvent was a combination of 70% (v/v) ethanol and 70% (v/v) methanol. Approximately 50 grams of plant powder were loaded into a cartridge along with 500 milliliters of the chosen solvent. Following a six-hour extraction period, the solvent was evacuated under vacuum, leaving behind a dry residue. This residue was subsequently recuperated, filtered using ashless filter paper (Whatman No. 4), and securely stored at a temperature of 4°C for subsequent analyses. The extraction was repeated three times to calculate the yield average. Formula (1) was used to compute the yield (%):

$$EY (\%) = \left(\frac{M_{ex}}{M_{pm}} \right) \times 100 \quad (1)$$

where: *EY* – extraction yield (%), *M_{ex}* – Mass of the extract (g), *M_{pm}* – mass of the plant material (g).

Total polyphenol assay

The quantification of total polyphenol content was conducted using the Folin-Ciocalteu (FC) method, following the procedure outlined by (Elbouzidi et al., 2023b). A mixture was prepared, consisting of 100 microliters of the olive extract, 500 microliters of FC reagent, and 400 microliters of a 7.5% (m/v) Na₂CO₃ solution. This mixture was vortexed and incubated in darkness at room temperature for 30 minutes. The absorbance

was measured at 760 nm employing a UV spectrophotometer (Perkin Elmer, Germany). The experiment was performed in triplicate. The results were expressed in milligrams of gallic acid equivalent (GAE) per gram of extract (mg GAE/g E) (Elbouzidi et al., 2023b).

Total flavonoid assay

The assessment of total flavonoid content in the extract was conducted via a colorimetric method based on the protocol established by (Elbouzidi et al., 2023a, 2022). Each sample (0.5 milliliters) was mixed with 2 mL of distilled water, followed by the addition of 0.15 mL of a 15% NaNO₂ solution. After 6 minutes, 0.15 mL of a 10% aluminum chloride (AlCl₃) solution was added and allowed to sit for an additional 6 minutes. Subsequently, 2 milliliters of a 4% NaOH solution was introduced into the mixture. The final volume was adjusted to 5 milliliters with distilled water, gently mixed, and placed in darkness at room temperature for 15 minutes. The absorbance was determined at 510 nm. Three repetitions of the test were conducted in order to reduce experimental error. The flavonoid content was expressed in milligrams of catechin equivalent per gram of dry matter (mg catechin/g dried extract).

Antioxidant activity assessment

To gauge the antioxidant potential of the extracts, we employed the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging method. This method largely followed the procedure outlined by (Haida et al., 2020; Hayani et al., 2022; Zroui et al., 2021), with minor adjustments. A solution of DPPH at a concentration of 76 μM (0.03 mg/mL) was prepared in ethanol. Subsequently, 2 mL of this DPPH solution was combined with 0.1 mL of various extract concentrations, ranging from 0 to 3 mg/mL. In parallel, a blank sample was prepared containing only the DPPH solution and methanol. After a 30-minute incubation period in darkness at room temperature, the absorbance of the reaction mixture was measured at 517 nm using a UV spectrophotometer (UV-2005, Selecta). Ascorbic acid served as the positive control in these experiments. The experiment was run through three times. The percentage inhibition of the DPPH radical was computed using the following formula 2:

$$\text{Inhibition (\%)} = \left(\frac{Ab - Ae}{Ab} \right) \times 100 \quad (2)$$

where: *Ab* – Absorbance of the blank, *Ae* – Absorbance of the sample.

The antioxidant activity was evaluated by determining the IC₅₀ value, which represents the concentration of either ascorbic acid or the extract that can scavenge 50% of the DPPH radical, as described by Tailor and Goyal (2014). The IC₅₀ value was calculated using the linear equation (*y* = *ax* + *b*) derived from the regression curve that correlates the percentage inhibition with the different concentrations of each sample:

$$IC_{50} = \left(\frac{50 - b}{a} \right) \times 100 \quad (3)$$

where: *a* – slope of the line, *b* – intercept at the origin.

Antibacterial evaluation

The antimicrobial effectiveness of the extracts was ascertained through the disk diffusion method, utilizing Muller Hinton Agar (MHA) medium. The methodology closely followed the guidelines provided by reference 16. The extracts were subjected to examination against six pathogenic bacterial strains, namely *Staphylococcus aureus*, *Escherichia coli*, *Acinetobacter baumannii*, *Salmonella* sp., *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*. The selection of these bacterial strains was based on their pathogenicity and their potential to contaminate food sources.

To initiate the assessment, isolated colonies of the bacterial strains were transferred to tubes containing sterile water. Subsequently, the surface of the MHA plates was inoculated with the microbial suspension. Sterile discs, each measuring 6 millimeters in diameter, were loaded with 10 μL of the extract, which had been appropriately diluted in dimethyl sulfoxide (DMSO) to achieve a concentration of 10 mg/mL, as outlined (Nounah et al., 2019). These discs were then strategically positioned on the surface of the MHA plates. The incubation of these plates was carried out at a temperature of 37°C for a duration of 24 hours, with the experimental tests conducted in triplicate.

The diameters of the inhibition zones, measured in millimeters, served as indicators of the antimicrobial activity of the extracts, while ceftazolin and erythromycin were employed as positive control substances.

Insecticidal impact

Insects and dose preparation

The insecticidal assessment was conducted using aphids that were cultivated under controlled laboratory conditions, maintaining a temperature of $25 \pm 2^\circ\text{C}$, a relative humidity level of 75%, and a photoperiod of 14 hours of light followed by 10 hours of darkness. The methodology involved infesting bean plants of the susceptible Defes variety, specifically at a less advanced vegetative stage marked by the fourth node, within insect breeding cages. To create various dosages, five different concentrations were prepared, with each concentration varying based on the dilution of the stock solution for each sample in distilled water. These doses were denoted as follows:

- Dose D1: 0.5%, corresponding to 0.05 grams of plant extract mixed with 10 mL of distilled water.
- Dose D2: 1%, equaling 0.1 grams of plant extract combined with 10 mL of distilled water.
- Dose D3: 1.5%, involving 0.15 grams of plant extract mixed with 10 mL of distilled water.
- Dose D4: 2%, consisting of 0.2 grams of plant extract blended with 10 mL of distilled water.
- Dose D5: 2.5%, represented by 0.25 grams of plant extract incorporated into 10 mL of distilled water.

Contact test

The examination of contact toxicity was conducted within controlled laboratory settings, maintaining a temperature of $25 \pm 2^\circ\text{C}$, a relative humidity of 75%, and adhering to a photoperiod of 14 hours of light followed by 10 hours of darkness. This evaluation involved the application of five different concentrations (0.5%, 1%, 1.5%, 2%, and 2.5%) for each plant extract. These concentrations were administered directly through the use of a 30 ml sprayer. To ensure the validity of the results, a completely randomized device was employed, with each concentration tested in five replicates. This device included a control group treated with water and a 0.01% Triton X-100 solution, which served as an emulsifier for blending distilled water with the extracts.

In the experimental setup for each concentration, five adult specimens were introduced into a glass petri dish. Each dish contained a piece of Whatman No.1 filter paper, as well as a fresh bean leaf provided as a source of nourishment.

This procedure was repeated five times for each concentration.

The assessment of mortality was carried out at distinct time intervals: one, two, three, 24, and 48 hours after the application of the extracts. This was done using a binocular microscope (Motic). To classify an individual as deceased, the assessment was based on the criterion that only those adults unable to move their legs were considered as having expired. The death rate was calculated utilizing the Abbot formula (4) as described in (Khandekar et al., 2023).

$$\text{Corrected mortality} = \frac{\%mortality\ in\ treatment - \%mortality\ in\ control}{100 - \%mortality\ in\ control} \times 100 \quad (4)$$

Statistical analysis

Subsequent to the data acquisition, a statistical analysis was executed employing the t-student test. The creation of graphical representations was undertaken using Microsoft Excel software (Version 2019), developed by Microsoft Corporation.

RESULTS AND DISCUSSION

Extraction yields using the soxhlet method

Table 1 presents the extraction yields obtained through the employment of the Soxhlet method. Notably, the methanol extract exhibited the highest yield, averaging at 24.3%, while the ethanol extract yielded the least, with an average of 14.3%. It is worth noting that previous studies have commonly reported higher yields when utilizing methanol as the solvent, a trend that aligns with our findings (Dinnies Santos et al., 2012). For context, our results are consistent with the findings of (Izza, 2020), which reported a methanolic extract yield of 36.75%, and (Salem and Saker, 2022), which documented a notable ethanol extract yield of 35.89%. However, it is essential to acknowledge that our results indicate

Table 1. Extract yield

Extracts	Yield %
Methanol extract	24.3 \pm 1.04 ^a
Ethanol extract	14.3 \pm 0.66 ^b

Note: data are presented as Mean \pm standard deviation (SD). each test was conducted in a triplicate. a, b different letters present a statistical difference at $p < 0.05$ using unpaired t-test.

comparatively lower yields in contrast to those reported by other authors. The variance in extraction yields can be attributed to several variables, including the specific plant species used in the extraction, the conditions under which the plant material was dried, the inherent metabolite content of each species, and the characteristics of the solvent employed for extraction or fractionation, taking into account its polarity (Ng et al., 2020). Furthermore, the divergence in outcomes can be linked to a range of factors, such as pH, temperature, extraction duration, and the composition of the sample. The timing of harvest and the geographical location of the plant material collection also exert a significant influence on the extraction yield (Madani Yousfi, 2017; Touaibia and Chaouch, 2014). Data were presented as Mean \pm standard deviation (SD). Each test was conducted in a triplicate. a, b different letters present a statistical difference at $p < 0.05$ using unpaired t-test.

Phenolic content analysis

The outcomes of the assessment of total phenolic and flavonoid content in the methanolic and ethanolic extracts of the investigated olive leaves (*Olea europaea* L.) are detailed in Table 2. Notably, these results underscore the richness of olive leaves in terms of total phenolics and flavonoids. Specifically, the methanolic extract reveals the highest content, with values measuring 185.64 ± 1.64 mg GAE/g DW for total phenols and 135.04 ± 0.76 mg RE/g DW for total flavonoids. In contrast, the ethanolic extract displays relatively lower levels, registering values of 149.06 ± 0.95 mg GAE/g DW for total phenols and 120.41 ± 1.35 mg RE/g DW for total flavonoids. These results become even more meaningful when compared to the findings of other researchers (Addab et al., 2020). It is evident that the variance in solvent polarity significantly influences the extraction of phenolic compounds. Additionally, the geographical region of harvest can also impact these results (Touaibia and Chaouch, 2014).

Table 2. Total content of phenols (TCP), Total content of flavonoids (TCF) of ethanolic and methanolic extracts of *Olea europaea* L.

Extracts	TCP (mg GAE/g E)	TCF (mg RE/g E)
Methanolic extract	185.64 ± 1.64^a	135.04 ± 0.76^a
Ethanolic extract	149.06 ± 0.95^b	120.41 ± 1.35^b

Note: data are presented as Mean \pm standard deviation (SD). each test was conducted in a triplicate. a, b different letters present a statistical difference at $p < 0.05$ using unpaired t-test.

Numerous investigations into the phenolic content of olive leaves have demonstrated the intricate interplay of various factors. These include the origin of the leaves, the drying process employed, the timing of harvest, the type of extraction solvent used, and the conditions under which the extracts are stored (Bolanle et al., 2014; Oubihi et al., 2020a; Touaibia and Chaouch, 2014). Furthermore, the qualitative and quantitative concentrations of total polyphenols are subject to variation between different plant species, a phenomenon often attributed to climatic and environmental factors (Zargoosh et al., 2019). Data were presented as mean \pm standard deviation (SD). Each test was conducted in a triplicate. a, b different letters present a statistical difference at $p < 0.05$ using unpaired t-test.

Antioxidant efficacy of olive leaves

Table 3 and Figure 1 provide a comprehensive depiction of the results pertaining to the percentage inhibition of the DPPH free radical by the methanolic and ethanolic extracts of *Olea europaea* L. These outcomes are juxtaposed with the inhibition percentages achieved by a potent antioxidant, ascorbic acid, which was employed as a positive control in this study. The IC_{50} value stands as a crucial metric inversely correlated with the antioxidant potential of a compound. Essentially, it quantifies the amount of antioxidant necessary to diminish the concentration of the

Table 3. Antioxidant activity of ethanolic and methanolic extracts of olive leaves

Extracts	DPPH (IC_{50} mg/mL)
Methanolic extract	0.426 ± 0.015^b
Ethanolic extract	0.711 ± 0.016^a
Acid ascorbic	0.090 ± 0.008^c

Note: data were presented as Mean \pm standard deviation (SD). each test was conducted in a triplicate. Different letters present a statistical difference at $p < 0.05$ using unpaired t-test.

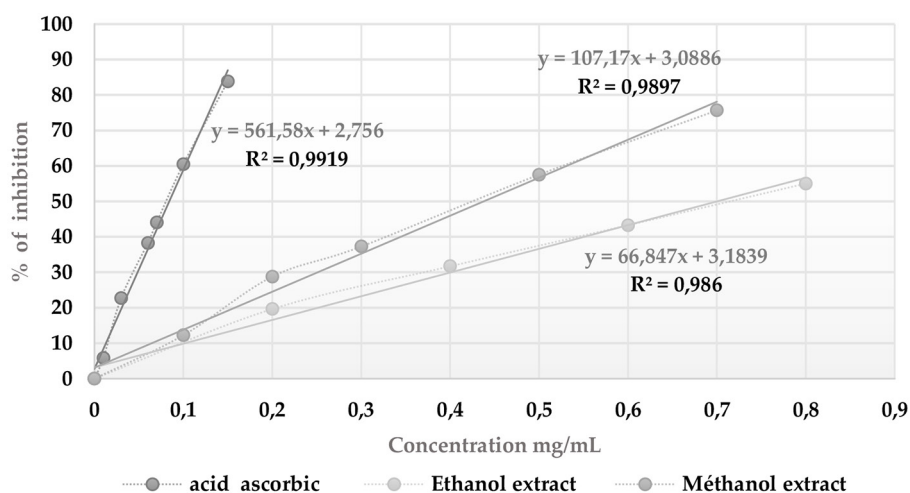


Figure 1. Dependence of the DPPH percentage inhibition on the concentration of methanol extract, ethanol extract and ascorbic acid

free radical by 50%. A lower IC_{50} value signifies a higher antioxidant capacity for the compound under examination (Izza, 2020).

Notably, the methanolic extract emerges as the most potent, exhibiting an IC_{50} value of 0.426 mg/mL, closely followed by the ethanolic extract with an IC_{50} value of 0.711 mg/mL. In contrast, ascorbic acid boasts a notably low IC_{50} value, measuring at 0.090 mg/mL, which aligns with its robust anti-radical prowess. Nevertheless, it is crucial to recognize that comparing these results with those of other studies is rendered inappropriate due to various factors influencing antioxidant content, including the choice of solvent and the adoption of different extraction techniques (Oubihi et al., 2020b). The observed significant anti-radical activity of these extracts against the DPPH radical can be primarily attributed to their inherent richness in phenolic compounds. Indeed, there exists a discernible correlation between the antioxidant effectiveness and the content of phenolic compounds. These findings are in concordance with the research of (Belhaddad, 2018), which highlighted that polar solvents prove to be optimal extraction media for phenolic compounds, especially flavonoids. This is primarily due to the presence of hydroxyl groups in their chemical structure, as exemplified by oleuropein.

However, it is essential to underscore that the antioxidant activity of plant extracts is contingent upon a multitude of variables. These include the composition of diverse antioxidants, the prevailing climatic conditions during plant growth, the stage of maturity at the time of harvest, storage conditions encompassing temperature and

duration, moisture content, pH levels, the type and polarity of the extraction solvent, the methodology employed for compound separation, the purity of bioactive substances, as well as the analytical techniques and the substrate utilized (MEDDOUR Elmoundir, 2021).

Assessment of antibacterial efficacy

The evaluation of antibacterial activity was conducted through the utilization of the disk diffusion method on solid medium. This qualitative technique revolves around measuring the diameter of bacterial growth inhibition in millimeters. Upon analyzing the results acquired from the measurement of the zones of inhibition produced by the methanolic and ethanolic extracts of olive leaves against the tested bacteria, it is evident, as presented in Table 4, that no discernible effect was observed. This suggests that the extracts did not exhibit antimicrobial activity against these pathogenic organisms. In fact, these outcomes were notably less pronounced compared to the inhibitory effects achieved by standard antibiotics. For instance, Cefazolin resulted in an inhibition diameter of approximately 12 mm against *Staphylococcus aureus*, while Erythromycin yielded inhibition diameters ranging from 9 mm to 16 mm for *Escherichia coli*, *Acinetobacter baumannii*, and *Klebsiella pneumoniae*.

It's important to note that when comparing our results with those of other studies, substantial disparities are apparent. One plausible explanation for these discrepancies may lie in the utilization of varying methods and distinct plant

Table 4. The antibacterial activity of olive leaves extracts by the diffusion method on disc.

Microorganisms	<i>Olea europaea</i> L.		Antibiotic	
	Methanolic extract	Ethanollic extract	Cefazolin	Erythromycin
<i>S. aureus</i>	00 ± 0.00	00 ± 0.00	12 ± 0.00	00 ± 0.00
<i>E. coli</i>	00 ± 0.00	00 ± 0.00	00 ± 0.00	09 ± 0.00
<i>A. baumannii</i>	00 ± 0.00	00 ± 0.00	00 ± 0.00	16 ± 0.00
<i>Salmonella sp</i>	00 ± 0.00	00 ± 0.00	00 ± 0.00	00 ± 0.00
<i>K. pneumoniae</i>	00 ± 0.00	00 ± 0.00	00 ± 0.00	09 ± 0.00
<i>P. aeruginosa</i>	00 ± 0.00	00 ± 0.00	00 ± 0.00	00 ± 0.00

Note: data are presented as Mean ± standard deviation (SD).

materials stemming from different olive varieties. Certain researchers have suggested a direct link between the chemical composition of phenolic elements and their antimicrobial activity. However, despite the richness of our extracts in polyphenols and flavonoids, no discernible antibacterial effects were observed. This phenomenon may be attributed to the intricacies of the cell walls of the targeted microorganisms, coupled with their sensitivity to the specific extracts (Miri and Djenane, 2019). The absence of antibacterial activity could be attributed to the potential resistance developed by certain bacterial strains, which may react differently to various extracts. These bacteria could inherently possess resistance due to the characteristics of their outer membrane (Jaber et al., 2021). It’s also worth noting that the diameter of the inhibition zone is influenced by several factors, encompassing the ability of substances present in the extracts to diffuse into the agar medium, the antimicrobial potency of these diffused substances, the growth rate, and metabolic activity of microorganisms within the medium (Jaber et al., 2018a; Oubihi et al., 2020c). Additionally, factors

such as the date of harvest may contribute to notable variations in the chemical composition and activity (Haddou et al., 2023). The determination of the inhibition zone diameter, including the 6 mm disc diameter, was accomplished through the agar disc diffusion method, utilizing a 15 µl volume of the extract disc and a concentration of 30 µg/disc for both Cefazolin and Erythromycin.

Assessment of the aphicidal activity

Figures 2 and 3 depict the cumulative and adjusted mortality rates observed following the treatment of *Aphis fabae* populations with varying concentrations of methanolic and ethanolic extracts olive leaves, with respect to time. The findings reveal a marked impact on the cumulative mortality of the insects when exposed to increasing doses of methanolic and ethanolic extracts of olive leaves. The effect is contingent upon the concentration employed and the duration of exposure. Notably, no mortality was observed within the control group. The methanolic extract exhibited a notable toxic effect on black

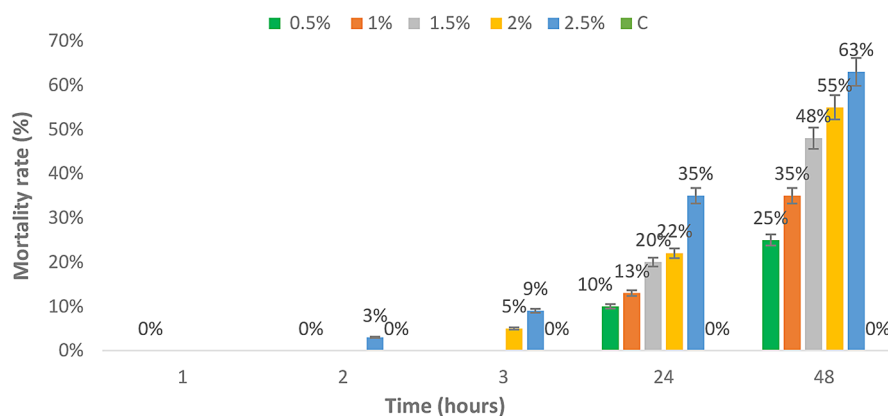


Figure 2. Effect of the methanolic extract of olive leaves on black bean aphid mortality. Results are presented as mean ± standard deviation

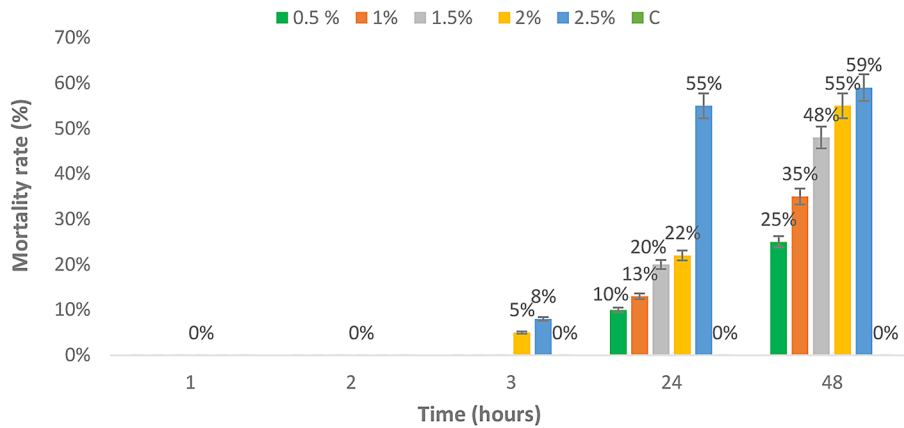


Figure 3. Effect of the ethanolic extract of olive leaves on black bean aphid mortality. Results are presented as mean \pm standard deviation

aphids, leading to a mortality rate of up to 63% on the second day of contact when exposed to a concentration of 2.5%. This effect was observed after just 2 hours. Similarly, the ethanolic extract induced a mortality rate of 59% on the second day when utilizing the same concentration. The absence of mortality in the control group strongly suggests that the observed mortality is primarily attributed to the insecticidal properties of the tested extracts. These findings align with the work of (Licir et al., 2021), who noted the considerable effectiveness of the aqueous extract of *Olea europaea* L. after just 4 hours of treatment at a 100% concentration. In contrast, Licir et al. (2021) reported lower mortality rates, commencing at 48 hours with a rate of 12.5% and reaching 25% after 72 hours for individuals treated with olive leaf extracts. The documented average mortality rate for olive leaves extracts can be attributed to the presence of secondary substances and natural phenolic compounds responsible for their anti-insect and repellent properties, as corroborated by the studies of Benyazza et al. (2021), and Hamouda et al. (2015).

The variability in the response of *Aphis fabae* to the treatment can be ascribed to multiple factors, including the chemical composition of the extracts, the nature of the solvent employed, application conditions, the extraction methodology, as well as the time and dose of treatment. Additionally, the stage of the pest's development, the season during which samples were collected, and the specific part of the aromatic plant tested are also influential variables. Attia and al. (2013), have pointed out that the effectiveness of plant extracts in insect control is still not well understood,

as they can exert physiological effects on insects, affecting their growth, molting, fecundity, and development. This study underscores that the plant extracts, regardless of their mode of application, contain active components with insecticidal properties, positioning them alongside other plants with similar characteristics that can potentially combat black aphids. It can be concluded that these extracts have the potential for inclusion in pest protection strategies.

CONCLUSIONS

In conclusion, *Olea europaea* L. leaves exhibit limited efficacy against bacterial strains; however, it boasts a substantial abundance of phenolic compounds, especially flavonoids. Furthermore, it demonstrates a robust capacity for counteracting cellular damage induced by free radicals. These characteristics position it as a top-tier antioxidant agent, opening avenues for therapeutic applications. It's important to note that antioxidants play a highly effective role in the prevention of various diseases like cancer and cardiovascular ailments. Additionally, its utility extends to enhancing food production and facilitating fragrance manufacturing. At the culmination of this study, it becomes evident that olive leaves' extracts undeniably possess insecticidal properties against insect species, particularly aphids. These extracts encompass chemicals recognized for their toxicity to these insects. The application of extracts from aromatic and medicinal plants to food surfaces proves to be a remarkably effective method for pest control. This approach offers numerous advantages

for both human health and the environment when compared to the use of chemically synthesized products. The present findings strongly suggest that the extract's combined antimicrobial and antioxidant activities are the result of multiple components working in concert, providing a solid rationale for its utilization in traditional medicine.

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