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Research Article

Investigating The Inhibitory Effects of Olive Oil on *Staphylococcus Aureus* and *Candida Albicans* Growth: A Comparative Study

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Abstract

Background: Extra virgin olive oil is a key ingredient in the Mediterranean diet, which is considered among the healthiest diets in the world. These benefits are attributed to its unique chemical composition, which consists mostly of oleic acid, in addition to bioactive phenolic substances such as hydroxytyrosol, tyrosol, and oleuropein. In this study, we investigated the antimicrobial activity of olive oil extracts against some pathogenic microorganisms.

Methodology: Biological activity was quantified using standard in vitro assays, including the agar diffusion method. Potato dextrose agar (PDA) was used as the culture medium to test the antifungal properties. Different concentrations of the substance (100%, 75%, and 50%) were cultured on inoculated plates. After an incubation period of 48–72 hours at 25–30°C, the diameter of the inhibition zones was measured in millimetres.

Results: The results showed a clear concentration-dependent antimicrobial effect. Candida showed the highest sensitivity, with inhibition zones of 28 mm, 25 mm, and 20 mm at 100%, 75%, and 50% concentrations, respectively. Staphylococcus (Gram-positive): Showed moderate sensitivity at high concentrations (15 mm at 100%), but exhibited resistance (R) at 50%.

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KEYWORDS: Olive oil, antimicrobial activity, Candida albicans, Staphylococcus aureus, Concentration-dependent inhibition.

1. INTRODUCTION

Natural products continue to provide new molecules and models for synthetic drug discovery, including anticancer agents [Stan et al., 2021]. Extra virgin olive oil has attracted much attention, given its antimicrobial effect against a wide spectrum of pathogens (Fancelo et al., 2022). This premium type of olive oil is extracted directly from olive fruits (Olea europaea) and cold-pressed without the use of chemicals, retaining most of its original bioactive components. This unique combination of antioxidants, monounsaturated fatty acids, and polyphenols makes extra virgin olive oil stand out from other vegetable oils and undoubtedly contributes to its health benefits. Fungal infections are a significant contributor to infectious disease-related mortality worldwide (Lee et al., 2021). Among these, Candida species are the most common causes of invasive mycotic disease, with C. albicans being the predominant agent of invasive candidiasis. In healthy individuals, Candida albicans is usually present as a harmless symbiotic organism in the oral cavity or gastrointestinal tract, except that in patients with severe immunodeficiency, this organism can move into the bloodstream, spread, and cause internal organ infection. Fatal systemic candidiasis. The fungal filaments are one of the main components that contribute to the hardness of this yeast. Here, the ability to transform from a yeast-like to a filamentous form is associated with increased rigidity and enhanced tissue-penetrating ability (see page no. (Lopez et al., 2022). Candida albicans are no longer malignant in their natural form, but they can become so as a result of changes in diet, antibiotic therapy, pH, or immune system disorders, where they multiply rapidly, causing symptoms and skin disorders. Vaginal, oropharyngeal, and intestinal candidiasis represented invasive infections in multiple organs (De Pipelands et al., 2022). Candidiasis is often associated with the presence of these biofilms on abiotic or biotic surfaces. Biomembranes are typically composed of compositionally complex communities of microorganisms embedded in a self-producing polymeric matrix (Atrial et al., 2021). Candida albicans fungus biofilms regularly release the characteristic long-layered yeast cells during the incubation period, facilitating the bacteria to form colonies at new sites of infection. This stage of diffusion ensured the repetition of the "life cycle of the molecule". Hepatitis is an important factor in the development of fungal infections, complicating treatment and increasing disease severity and cell death. Increased resistance of biomembrane-embedded cells to antifungal drug therapy, and their presence in the host immune system, are the two major clinical problems associated with biofilm formation (Pereira et al., 2021).

Only a few types of antifungal drugs are available for the treatment of this potentially fatal disease, despite the negative impact of fungi on human health. The nature of fungal cells containing whole nuclei and the difficulty of drug binding to the fungal cell wall and its membrane may have contributed to the slow clearance of antifungal drugs (Fan et al., 2021).

2. MATERIALS AND METHODS

Experimental Procedure for Detecting Biological Activity

Materials and Methods

2.1 Study Design

An experimental laboratory study was applied to investigate the antimicrobial activity of olive oil against bacterial and fungal isolates.

2.2 Collection of Microbial Isolates

Seven isolates of *Staphylococcus aureus* and 7 isolates of *Candida albicans* were used in this study. The isolates were obtained from laboratory cultures and maintained under appropriate microbiological conditions.

2.3 Preparation of Olive Oil Concentrations

Three concentrations of olive oil were prepared for antimicrobial testing:
100% olive oil, 75% olive oil, and 50% olive oil
Sterile distilled water was used as a control

2.4 Culture Media

The following culture media were used: Mueller-Hinton agar for bacterial isolates (Atlas et al., 2010). Sabouraud dextrose agar for fungal isolates. The media were prepared according to the manufacturer's instructions and sterilised by autoclaving.

2.5 Preparation of Microbial Suspensions

Bacterial and fungal suspensions were prepared from fresh cultures and adjusted to approximately 0.5 McFarland standard to ensure uniform microbial concentration (14,13).

2.6 Agar Well Diffusion Method

The antimicrobial activity of olive oil was determined using the agar well diffusion method. Sterile swabs were used to inoculate the microbial suspensions onto agar plates. Wells were made in the agar using a sterile corn borer. Different concentrations of olive oil were added to the wells. The plates inoculated with *Staphylococcus aureus* were incubated at 37°C for 24 hours, while plates inoculated with *Candida albicans* were incubated at 30°C for 24–48 hours. After incubation, inhibition zones around the wells were measured in millimetres.

2.7 Microorganisms

14 isolates of Staphylococcus aureus and Candida fungi were obtained from Al-Amin Centre for Scientific Research in Najaf.

2.8 Disc Diffusion Method Procedure

Therefore, the presence of inorganic oil was evaluated to evaluate its effectiveness as a germicide. Using a cotton swab, the bacterial suspension was spread on Mueller-Hinton agar plates after adjusting to 0.5 of the McFarland scale. A sterile filter paper disk with a diameter of 6 mm was then used, and 10 µL of herbal oil at the desired concentration was placed on each disk. Three concentrations were prepared for each extract:

second concentration: 100, 75, and 50 µL of crude oil. Subsequently, the agar plates were incubated in an incubator for 24 h at 37 °C. After incubation, the diameter of the zone of inhibition was measured to evaluate the efficacy of the herbicide (Forbes et al., 2005). The results of these measurements represented the inhibition area of the crude oil.

2.9 Statistical Analysis

The size of the inhibitory zone was measured and compared between the tested force levels. and the size of the infarct area in millimetres was used to interpret the results. The biological activator is the first to emerge from the standardised analysis conforming to the laboratory. Certainly, there is a mitigating procedure, such as the use of antimicrobial/antimicrobial protection. Effectiveness was then assessed, and the antimicrobial.

The effect was tested by a measurement of the inhibition zone). The outcomes thereof are the inhibition zones of olive oil in Table 1 and Figure 1.

3. RESULT AND DISCUSSION

3.1 used Olive Oil as an Antibacterial Activity Against Staphylococcus aureus

The Staph. aureus isolates against the activity of olive oil are presented in Figure 1 and Table 1. The results showed that the 100% concentration appeared to have the highest antibacterial activity, with a reduction in the range of zones from 10 to 16 mm. The 75% concentration showed moderate antibacterial activity with inhibition zones ranging from 10–14 mm. However, the 50% concentration showed no inhibitory activity against all tested bacterial isolates (Prescott,2005).

Figure 1 shows the inhibition zone by olive oil against Staph. arous isolate

Table 1. Inhibitory diameters of olive oil against Staphylococcus aureus

Sample	100	75	50	Control
S1	15	13	R	0
S2	12	10	R	0
S3	16	12	R	0
S4	12	14	R	0
S5	13	10	R	0
S6	10	12	R	0
S7	14	13	R	0

R=Resistance

The results indicate that increasing olive oil concentration enhanced the antibacterial activity against Staphylococcus aureus.

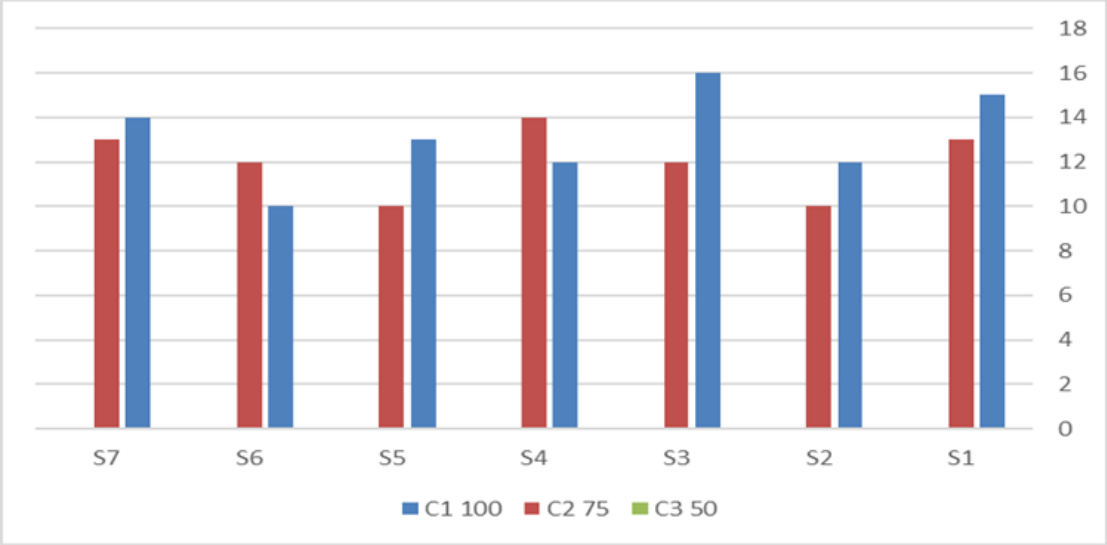

Figure 3: Effect of concentration of olive oil on the isolates of Staph.

3.2 Antifungal Activity of Olive Oil Against Candida albicans

The antifungal activity of olive oil toward Candida albicans isolated is shown in Figure 2 and Table 2. Strong antifungal activity was shown in information at all tested doses. The greatest inhibitory zones, measuring between 24 and 30 mm, were seen at 100% concentration. With inhibition zones spanning from 22 to 28 mm, the 75% concentration also showed strong antifungal action in Figure 3. All fungal isolates were moderately inhibited by the 50% concentration

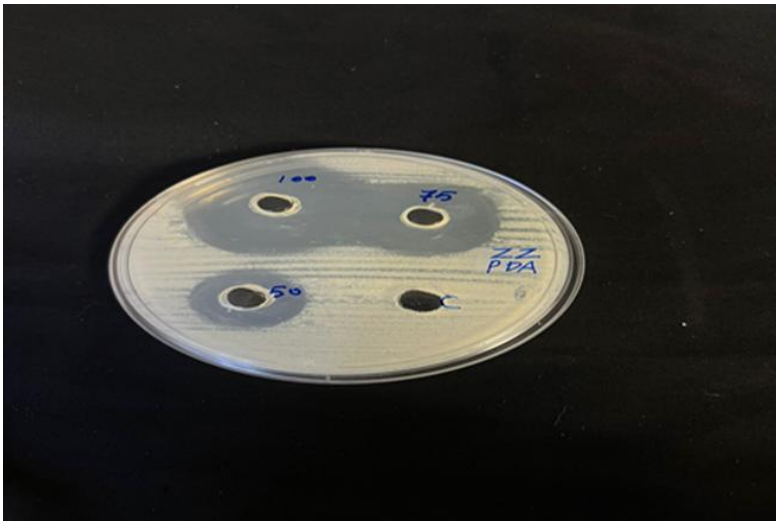

Figure 2: Candida cultured on an agar plate with olive oil with a zone of inhibition

Table 2. Inhibitory diameters of olive oil against Candida albicans

Sample	100	75	50	Control
CA1	28	25	20	0
CA2	25	27	21	0
CA3	25	24	20	0
CA4	30	22	23	0
CA5	24	24	22	0
CA6	26	26	20	0
CA7	27	28	21	0

These findings indicate that olive oil possesses greater antifungal activity than antibacterial activity.

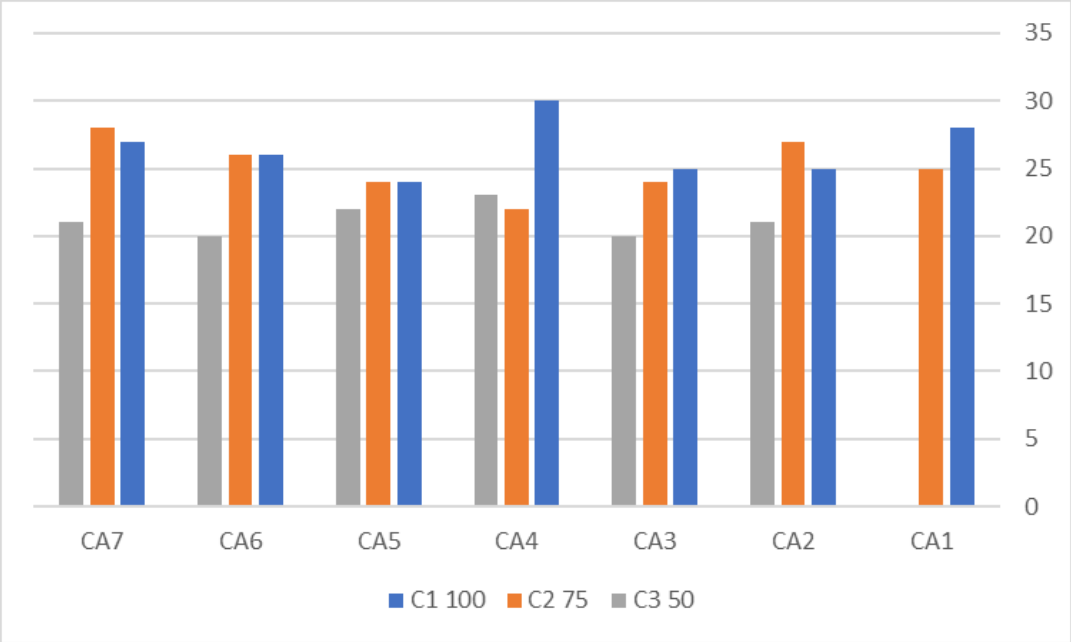

Figure 3: Effect Of Concentration of Olive Oil on The Isolates of Candida

4. DISCUSSION

Recent examination showed that olive oil has antimicrobial effects against Candida albicans and Staphylococcus aureus (Al-Waili,2005). However, the antibacterial activity against Staphylococcus aureus was lower than the antifungal effect against Candida albicans. The findings proved that the inhibition effect could be enhanced by raising the content of olive oil. At 100% concentration, the greatest inhibitory zones were observed, whilst lower values showed decreased activity. Bacterial cells may need larger concentrations of olive oil phenolics to perform an antimicrobial effect, as proven by the lack of inhibition against bacterial isolates at 50% concentration (Medina,2006).

Olive oil's phenolic components, including hydroxytyrosol, oleuropein, and tyrosol, may be the source of its antimicrobial effects. These substances can interfere with vital cellular functions and change the permeability of the membranes (Omar,2010). The sensitivity of fungal cell membranes to fatty acids and phenolic compounds found in olive oil may be the cause of the more susceptible nature of Candida albicans found in the present study. Components of olive oil may disrupt membrane integrity and ergosterol fabrication, which would impede the development of fungi (Silva et al.,2012) . The results of this investigation support earlier research showing that olive oil has an antibacterial effect against fungi and Gram-positive bacteria. Olive oil phenolics have been shown by Medina et al.2006) to have bactericidal effects against a variety of pathogenic bacteria, including Staphylococcus aureus. Similarly, combinations including olive oil had inhibitory effects against Candida albicans and Staphylococcus aureus, according to Al-Waili (2005). In accordance with other studies, the phenolic chemicals found in olive oil have potent antioxidant and antibacterial effects. Fungi and bacteria have different cell wall compositions, which might contribute to the more efficient protection against Candida albicans seen in this study. The membranes of fungal cells are rich in ergosterol and may be more susceptible to lipid-soluble materials. Genetic and physiological variations across microbial strains may be observed in the variation in inhibiting zones among isolates (Madigan,2012). Resistance mechanisms that lessen sensitivity to antimicrobial drugs may be present in some isolates. As demonstrated in the current study, olive oil can be used as a natural antibacterial. Because they may lessen the negative effects of synthetic antimicrobial medications and may aid in the fight against antimicrobial resistance, natural products receive more and more attention. Even though olive oil demonstrated encouraging anti-microbial action, more research is needed to determine the active elements causing the antimicrobial impact. Future research should look into how olive oil works in concert with antibiotics and antifungal medications (Karygianni,2016).

5. CONCLUSION

The present study demonstrated that olive oil possesses significant antimicrobial activity against Staphylococcus aureus and Candida albicans. The antifungal activity against Candida albicans was stronger than the antibacterial activity against Staphylococcus aureus. The highest inhibitory activity was observed at 100% concentration, while lower concentrations showed reduced effectiveness. Olive oil may represent a promising natural antimicrobial agent due to its richness in phenolic compounds and bioactive constituents. Plethora studies are recommended to discuss the reactivity of materials sensitivity for the antimicrobial activity and to measure the medical applications of olive oil in infection control.

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