

# Antimicrobial And Antioxidant Potentials Of Some Essential Oils Against Spoilage Microorganisms

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## Abstract

This study evaluates the antioxidant and antimicrobial activities of essential oils (EOs) from four Lamiaceae species: thyme, rosemary, oregano, and sage. Antioxidant potential was assessed through various assays, including DPPH radical scavenging, reducing power activity (RPA), and total antioxidant capacity (TAC). Antimicrobial activity was tested against five pathogenic bacteria (*Staphylococcus aureus*, *Bacillus cereus*, *Listeria innocua*, *Pseudomonas aeruginosa*, and *Escherichia coli*) and a yeast strain (*Candida albicans*). The results of the three antioxidant activity tests indicated that the potency of antioxidants increases in direct proportion to the concentration of essential oils. Oregano EO demonstrated the strongest DPPH scavenging activity and reducing power, surpassing standard antioxidants like ascorbic acid. Therefore, oregano and thyme EOs exhibited excellent antimicrobial properties against microbial strains tested, outperforming rosemary and sage EOs. These findings indicate that oregano and thyme essential oils possess considerable promise as natural preservatives in food and pharmaceutical applications, which may lead to the replacement of synthetic preservatives with plant essential oils.

**Keywords:** Essential oil, Lamiaceae, antibacterial activity, antifungal activity, antioxidant activity.

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## 1. INTRODUCTION

For thousands of years and over the centuries, aromatic and therapeutic herbs have been essential in human nutrition and medicine. Their traditional use as natural remedies is based on their richness in bioactive compounds with antimicrobial, antioxidant and curative properties (Cock et al., 2018). The complex natural components known as essential oils (EO) are extracted or distilled from plant materials. They are utilized in a variety of industries, including natural therapy, the pharmaceutical sector, and food preservation, because of its volatile components, which include thymol, carvacrol, and linalool (Guldiken et al., 2018; Kosakowska et al., 2021). The Lamiaceae family, which includes plants such as thyme, oregano, sage and rosemary, is widely recognized for its diverse biological properties (Lee et al., 2020; Grigoriadou et al., 2020; Aziz et al., 2022). These plants are an important source of antimicrobial and antioxidant compounds, making them particularly interesting for research (Çelik et al., 2021). With the growing resistance of microorganisms to antibiotics and concerns about synthetic additives, it is crucial to explore natural solutions. Essential oils from Lamiaceae offer a promising alternative for food bio-preservation and the fight against human pathogens (Walasek-Janusz et al., 2024). Our study aimed to evaluate *in vitro* the antimicrobial and antioxidant activity of four essential oils from Lamiaceae (thyme, oregano, rosemary and sage) against pathogenic microbial strains. The results of this study highlight the promising role of essential oils as biopreservatives, considered a good alternative to synthetic preservatives.

## 2. METHODOLOGY

### 2.1. Essential oils

Thyme, rosemary, oregano, and sage essential oils (EO's) were purchased from Voshuiles.com. The essentials oils were certified by Ecocert FR-BIO 01 (France).

## **2.2. Chemicals, reagents and standards**

The analytical grade, highest commercially available purity chemicals, solvents, reagents, and reference standards used were all acquired from Sigma-Aldrich in Germany. These included 2,2-diphenyl-1-picrylhydrazyl radical (DPPH), butylated hydroxytoluene (BHT), ascorbic acid, trichloroacetic acid, potassium ferricyanide, sodium phosphate, ammonium molybdate, dimethyl sulphoxide (DMSO), iron (III) chloride (FeCl<sub>3</sub>), and methanol. Liofilchem s.r.l., Italy, provided the 0.5 McFarland.

## **2.3. Antioxidant assays**

### **2.3.1. DPPH radical scavenging activity**

The Blois (1958) method was used to determine the stable DPPH radical scavenging activity. 1.0 mL of DPPH methanolic solution (0.2 mM) was combined with 500 µL of samples at various methanol concentrations. After mixing by vigorous shaking, the samples were incubated for 30 min at room temperature. The absorbance was measured at 517 nm using methanol as a blank in a UV-Vis spectrophotometer (Beckman; DU520). The following formula was used to determine the radical scavenging activity (RSA): % RSA = [(A control - A sample) / A control] x 100, where A sample is the absorbance of the samples/references and A control is the absorbance of the control reaction (which contains all reagents except the test sample). The graph of RSA percentage against oil concentration was used to determine the concentration that provided 50% inhibition (IC 50). For reference, the RSA of BHT and ascorbic acid were also calculated. The ascorbic acid equivalents (mg AAE/g) of EO were used to express the results.

### **2.3.2. Reducing power activity (RPA)**

Reducing power activity (RPA) of EO's was estimated using the Oyaizu (1986)'s approach. 2.0 mL of samples (10 mg/mL), 1.0 mL of phosphate buffer (2 mM, pH 6.6) and 1.0 mL of potassium ferricyanide [K<sub>3</sub>Fe(CN)<sub>6</sub>] were mixed and incubated for 20 min at 50 °C, then a 1.0 mL aliquot of the reaction mixture was removed after the addition of 1.0 mL of 10 % TCA, the sample was mixed with 1.0 mL of distilled water and 0.2 mL of 0.1 % FeCl<sub>3</sub>. A UV spectrophotometer (Beckman, DU520) was used to measure the absorbance of the chromogen at 700 nm against a methanol blank following a 10-minute incubation period at room temperature. High reducing power was linked to high absorbance levels. For contrast the reducing power of BHT and ascorbic acid was also calculated.

### **2.3.3. Total antioxidant capacity (TAC)**

EOs' total antioxidant activity (TAC) was measured using the methodology outlined by Hmamou et al. (2022). 1.0 mL of molybdate reagent, which contained 28 mM sodium phosphate, 4 mM ammonium molybdate, and 6 mM sulfuric acid, was mixed with 100 µL of samples. After 90 minutes of incubation at 95 °C, the mixture was allowed to cool to room temperature. A UV spectrophotometer (Beckman, DU520) was used to measure absorbance at 695 nm against a methanol blank.

## **2.4. Antibacterial assay**

### **2.4.1. Microbial strains**

Three Gram-positive bacteria (Staphylococcus aureus ATCC 25923, Bacillus cereus ATCC 14579, Listeria innocua ATCC 33090), two Gram-negative bacteria (Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 25922), and a yeast strain (C. albicans ATCC 10231) were used to evaluate the antimicrobial activity of EOs.

### **2.4.2. Disc diffusion method**

A densitometer was used to standardize bacterial suspensions made from early cultures (18–24 hours) at 0.5 McFarland, which were then spread out over Muller Hinton (MH) agar medium. The surface of the inoculated agars was covered with sterile paper discs (6 mm in diameter, Whatman no. 1) that had been sterilized by filtration and impregnated with 5 µL of EO diluted in a sterile DMSO to a concentration of 100 mg/mL. Amoxicillin (AX; 25 µg/mL), Oxacillin (OX; 1 µg/mL), Gentamicin (GN; 120 µg/mL), and Tetracycline (TC; 30 µg/mL) discs were used as the positive controls, while discs prepared with the same volume of methanol were used as negative controls. The diameter of the inhibition zones against the studied microorganisms was used to measure the antibacterial activity following a 24-hour incubation period at 37 °C. A scale for estimating antimicrobial activity published by Ponce et al. (2003) is used to interpret the results. They divided the diameters of the inhibition zones (DIZ) of microbial growth into four classes as follows: not sensitive for diameters under 8 mm, sensitive for diameters between 9 and 14

mm, very sensitive for diameters between 15 and 19 mm, and extremely sensitive for diameters over 20 mm.

#### **2.4.3. Micro-atmospheres**

Sterile Petri dishes containing (18 mL) MH agar inoculated with bacterial suspension were prepared. The dish lids were centered with discs (6 mm in diameter) that had been impregnated with 5  $\mu$ L of pure HE. For 24 hours for bacterial strains and 48 hours for yeast, the plates were then incubated at 37°C. Lastly, a caliper was used to measure the diameter of the zones of inhibition (Tao et al., 2021).

#### **2.4.4. Minimal inhibitory concentration**

The Mueller Hinton broth (MHB) dilution method, as defined by May et al. (2000), was used to calculate the minimal inhibitory concentration (MIC) of EOs. 5  $\mu$ L of a standardized bacterial culture (0.5 McFarland) was added to MHB tubes that contained varying concentrations of EO (0.000–1.000%, v/v). Every tube was incubated for 24 hours at 37 °C while being shaken at 120 rpm. At the same time, control tubes made using the same process but without the bacterial inoculum were evaluated. The lowest concentration that prevented discernible growth was represented by the MIC.

#### **2.5. Statistical analysis**

From triplicate data, means and standard deviations were calculated. Duncan's post-hoc test was used after a one-way ANOVA to examine differences at p-value < 0.05.

### **3. RESULTS AND DISCUSSION**

#### **3.1. Antioxidant activity**

##### **3.1.1. Kinetic study of the reaction by the DPPH test**

The percentage of inhibition of the DPPH radical was examined for different concentrations for each of the EOs studied and was compared with reference antioxidants, namely AA and BHT. The absorbance measurement was carried out by spectrophotometry at 517 nm, and from the results obtained, the inhibition percentages were calculated. Plotting the curves showing the fluctuation of the inhibition % as a function of the concentrations of thyme, rosemary, oregano, sage, AA, and BHT essential oils was made possible by the data obtained (Figure 1). From the results obtained, it is clear that, for both the reference standards (AA and BHT) and the several EOs examined, the percentage of inhibition of the free radical DPPH rises as concentration increases. However, the essential oils of sage and oregano have strong anti-radical properties against the free radical DPPH. (Table 1). The EO of oregano represents the most powerful antioxidant with a percentage of radical inhibition higher than that of AA (98.75% and 89.26%, respectively). The EO of sage has a high anti-radical activity (81.58%) compared to those of the EOs of thyme and rosemary, which reveal similar low anti-radical powers (17.9% and 20.89%, respectively). The concentration of EOs and reference standards causing 50% inhibition or reduction of DPPH (IC<sub>50</sub>) is calculated from the formula extracted from the trend curve of the percentage of inhibition as a function of the concentration for each compound (Figure 2). Referring to the IC<sub>50</sub>, oregano EO has a lower value than BHT and AA (37.60  $\mu$ g). The IC<sub>50</sub> recorded for sage EO is higher than that of BHT and close to that of AA. However, thyme and rosemary EOs have higher IC<sub>50</sub> values that exceed 1000  $\mu$ g (4455.05 and 2168.60  $\mu$ g, respectively). The amount of antioxidant needed to reduce the concentration of the free radical by 50% is expressed by IC<sub>50</sub>, which is inversely correlated with a compound's antioxidant capacity. Stated differently, as a substance's antioxidant activity increases, its IC<sub>50</sub> value decreases (Ismaili et al., 2017). Volatile terpenoids and polyphenolic compounds are the two main categories of secondary metabolites that have been linked to antioxidant activity among the bioactive chemicals extracted from sage (Grigore-Gurgu et al., 2025). Therefore, it is evident that oregano essential oil's phenolic components give it strong antioxidant properties (Sidiropoulou, 2022).

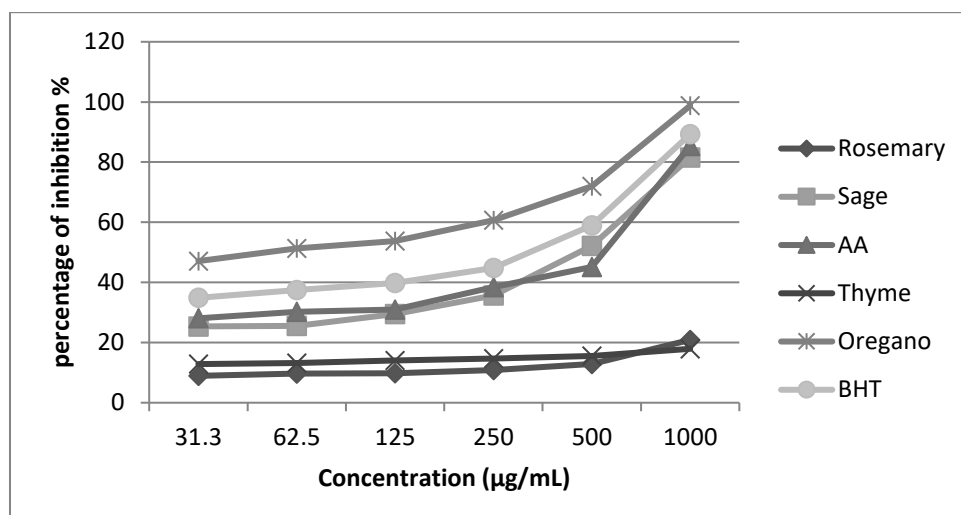


Figure 1: Percentage of DPPH inhibition as a function of the concentrations of EOs, BHT and AA.

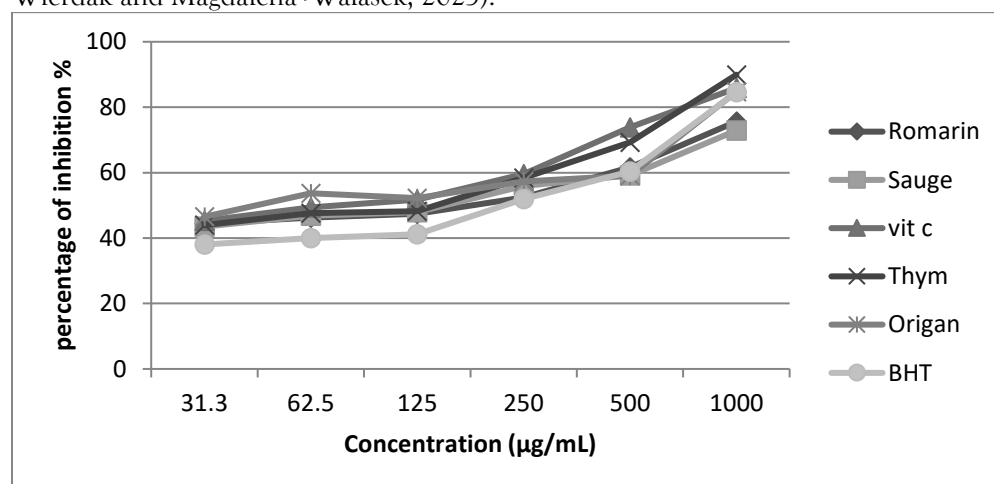
Table 1: IC<sub>50</sub> values for DPPH, RPA, and TAC antioxidant activity tests expressed in (µg/mL)

EO/ Standards	Antioxidant tests	DPPH/ RPA/ TAC % at the concentration of 1 mg/mL	IC <sub>50</sub> (µg/mL)
AA	DPPH	85.35	542.6
	RPA	85.87	66.30
	TAC	95.78	89.50
BHT	DPPH	89.26	354.22
	RPA	84.58	322.80
	TAC	74.25	213.90
Sage	DPPH	81.58	571.64
	RPA	72.91	188.00
	TAC	96.62	106.20
Oregano	DPPH	98.75	37.6
	RPA	84.74	60.01
	TAC	96.52	58.40
Rosemary	DPPH	20.89	2168.6
	RPA	75.62	218.24
	TAC	97.68	133.20
Thyme	DPPH	17.92	4455.05
	RPA	89.93	135.20
	TAC	98.73	104.40

### 3.1.2. Determination of reducing power activity (RPA)

The chemical reduction of ferric ions Fe (III) in the potassium ferricyanide complex  $K_3Fe(CN)_6$  into ferrous ions Fe (II) is the basis of the RPA method (SHAHIDI et al., 2018). The investigated Eos' reduction power activity was contrasted with that of the reference antioxidants, BHT and AA. The acquired results demonstrate that the increase in our Eos' concentration is correlated with the decreasing power activity (Figure 2). Numerous investigators have confirmed this fact. According to the results, Eos from sage and rosemary have a moderate capacity to lower iron, which is lower than that of AA and BHT. Compared to oregano essential oil, the latter exhibits a reducing power of 84.74%. 1,8-cineole,  $\alpha$ -pinene, camphor, borneol, camphene, p-cymene,  $\beta$ -pinene, and limonene are all present in different levels in rosemary essential oil. Alvarez et al. (2019) have proposed that rosemary's lipophilic components contribute less to its antioxidant action. Nevertheless, thyme EO has a greater ability to reduce iron than AA (89.93%). According to Table 1, the antioxidant BHT has the highest IC<sub>50</sub> value (322.80 µg). In contrast, oregano EO shows the lowest IC<sub>50</sub> value, close to that of AA (60.01 and 66.30 µg, respectively). Whereas the

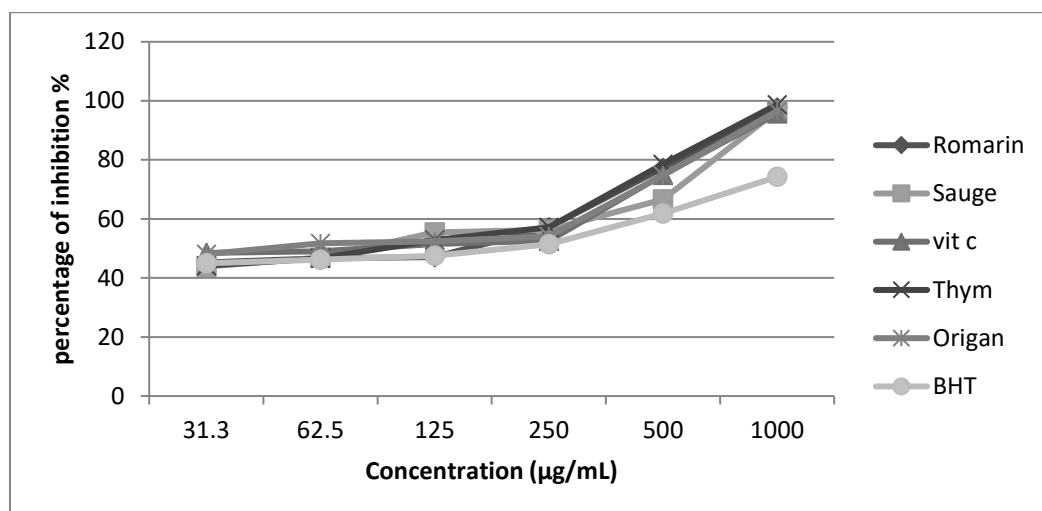
average levels of Eos for thyme, sage, and rosemary are 135.20, 188.006, and 218.24  $\mu\text{g}$ , respectively. A high concentration of thymol, carvacrol, or both is linked to essential oil activity (Kosakowska et al., 2021). Like thymol, carvacrol has the ability to scavenge free radicals and transform them into stable forms devoid of radical activity (Imran et al., 2022). Both of these compounds can donate hydrogen from the hydroxyl group connected to the aromatic ring, which turns free radicals into inactive forms. This is because they both have an aromatic ring, which lowers their reducing potential compared to free radicals (Walasek-Janusz et al., 2024). Consequently, they act as donors of hydrogen or electrons, which permits the reduction of  $\text{Fe}^{3+}$  and the conversion of the DPPH $\cdot$  radical into its reduced form, DPPH $\cdot$  (Nurzynska-Wierdak and Magdalena -Walasek, 2025).



**Figure 2:** Percentage of RPA inhibition as a function of the concentrations of EOs, BHT and AA.

### 3.1.3. Total Antioxidant Capacity (TAC)

To determine the overall antioxidant capacity, the phosphomolybdenum method is commonly used, simple, rapid, and independent (Dutta et al., 2012). Figure 3 shows a graphic representation of the total antioxidant capacity of the examined Eos and the standards (AA, BHT). The Eos of thyme, rosemary, sage, and oregano have TACs also higher than that of AA (98.73, 97.68, 96.62, and 96.52%, respectively). In contrast, BHT shows the lowest total antioxidant capacity (74%). The EO of oregano has the lowest IC<sub>50</sub> value (58.40  $\mu\text{g}$ ). Additionally, lower than AA, this exhibits an IC<sub>50</sub> value lower than that of Eos from thyme and sage. The latter exhibit similar IC<sub>50</sub> values (104.40 and 106.20  $\mu\text{g}$ , respectively). It is clearly established that rosemary EO has the highest IC<sub>50</sub> value (133.20  $\mu\text{g}$ ). However, BHT has the highest IC<sub>50</sub> value (213.90 $\mu\text{g}$ ) (Table 1). Large levels of monoterpenes with considerable antioxidant activity, including thymol,  $\gamma$ -terpinene, p-cymene, 1,8-cineole,  $\alpha$ -pinene, carvacrol, and linalool, as well as the sesquiterpene caryophyllene oxide, are found in the essential oils of thyme, oregano, rosemary, and sage. The overall phenolic content of herbs and their antioxidant qualities, however, were found to be linearly related. The antioxidant activity of natural resources is constantly studied, and several aromatic plants, including oregano species, have given promising results (Aebisher et al., 2021; Nurzynska-Wierdak and Magdalena -Walasek, 2025). In line with the research conducted by MECHERGUI et al. (2010), with an IC<sub>50</sub> value of 1280  $\mu\text{g}/\text{mL}$ , oregano EO demonstrated antioxidant activity by trapping the DPPH radical. This result is significantly higher than that recorded in our study. In contradiction, QUE et al. (2006) reported a lower IC<sub>50</sub> value for oregano EO of 2.80  $\mu\text{g}/\text{mL}$ . Comparing our findings with those of other previous research, we note that the IC<sub>50</sub> for thyme EO is greater than some of the values found in prior studies. Indeed, the work carried out by Ceylan et al. (2016) indicates that thyme EO has a remarkable efficiency of around 850  $\mu\text{g}/\text{mL}$ . On the other hand, our IC<sub>50</sub> appears close to that reported in Ismaili et al. (2017), who reported an IC<sub>50</sub> of around 4.57 mg/mL (4570  $\mu\text{g}/\text{mL}$ ), which for them indicates a low antioxidant activity compared to the other thymus species studied. Med Raâfet Ben Khedher et al. (2017) noted a higher result and recorded IC<sub>50</sub> values of sage EO of 6700  $\mu\text{g}/\text{mL}$  and 28400  $\mu\text{g}/\text{mL}$  for the DPPH and RPA tests, respectively.



**Figure 3:** Percentage of TAC inhibition as a function of the concentrations of EOs, BHT and AA.

### 3.2. Antimicrobial activity

#### 3.2.1. Disk diffusion method

By measuring the sensitivity of the various microbial species, the disk diffusion method was used to assess the antibacterial activity of the specified essential oils. The results of the evaluation of the antimicrobial activity of the different EOs studied on the different microbial strains tested are shown in Table 2 and Figure 4. Significant antibacterial activity was demonstrated by the essential oils of thyme, and oregano against every microbial strain that was examined with DIZ ranging from  $69.5 \pm 0.5$  to  $70.5 \pm 0.5$  mm, and  $32.00 \pm 1.0$  to  $77.05 \pm 0.5$  mm, respectively. Phenolic chemicals, which may stop bacterial growth even at the lowest dose, were primarily linked to the antibacterial properties of thyme and oregano essential oils (EOs) (Ibáñez et al., 2020; Mutlu-Ingok et al., 2021). Moreover, oregano oils' capacity to disrupt numerous essential bacterial processes, such as the formation of cell walls, proteins, and nucleic acids, accounts for their microbiological activity (Janusz and Walesek, 2024). *L. innocua*, *E. coli*, and *Candida albicans* are not susceptible to the antibacterial properties of rosemary essential oil with DIZ varied from  $06.00 \pm 0.00$  to  $08.00 \pm 1.00$  mm. This essential oil exhibited a modest level of antibacterial activity against *P. aeruginosa*, *B. cereus*, and *S. aureus*. Since Gram-positive bacteria do not have an outer membrane (OM), unlike Gram-negative bacteria, EOs often have a stronger effect on Gram-positive bacteria than on Gram-negative bacteria. This is likely because the two types of bacteria have different cell wall compositions (Khameneh et al., 2019). Sage essential oil had no antibacterial action against *P. aeruginosa*, *B. cereus*, or *L. innocua*. Moreover, this essential oil demonstrated strong antifungal activity against *Candida albicans*. However, it demonstrated outstanding antibacterial action against *S. aureus* and *E. coli* with DIZ of  $15.50 \pm 1.5$  mm, and  $61.0 \pm 1.00$  mm, respectively. Because of changes in the quality of the herbs, the qualitative and quantitative variations in the contents of essential oils, the discrepancies between the fungal strains that were studied, and methodological variations, the results of the many research regarding the antifungal potential are not always similar (Hammoudi Halat et al., 2022). Therefore, in contrast to the use of conventional antibiotics, we found that all bacterial strains were extremely sensitive to GN antibiotic with DIZ ranging from  $28.0 \pm 0.0$  to  $32.0 \pm 1.0$  mm. Additionally, *B. cereus* and *L. innocua* were extremely sensitive to AX with DIZ of  $26.0 \pm 0.0$  mm, and  $33,5 \pm 0.7$  mm; respectively. *P. aeruginosa*, *C. albicans*, and *E. coli* were resistant to both AX and OX. However, *S. aureus* was resistant to TC, and sensible to AX and OX. Wherefore, The World Health Organization has identified the rise of multi-resistant bacteria as a hazard to global health security (Kebede et al., 2021). The literature has documented the antibacterial properties of the various EOs that were investigated against the strains that were tested. Excellent antibacterial activity was noted for the tested EOs in Imane et al. (2020) and Walesek-Janusz (2024), who investigated the antimicrobial activity of thyme and oregano essential oils against the same tested strains. Bouras et al. (2019) examined the bacterial activity of oregano EO against some tested strains and revealed that this EO shown moderate bacterial activity against *B. cereus* and *E. coli* and high bacterial activity against *S. aureus*. The research conducted by Mouas et al. (2017) revealed that essential oil of rosemary has

moderate bacterial activity against *B. cereus* and *E. coli*, high bacterial activity against *S. aureus*, and no bacterial activity against *P. aeruginosa*.

**Table 2:** Sensitivity of microbial strains to the tested Eos and antibiotics (Disk diffusion method)

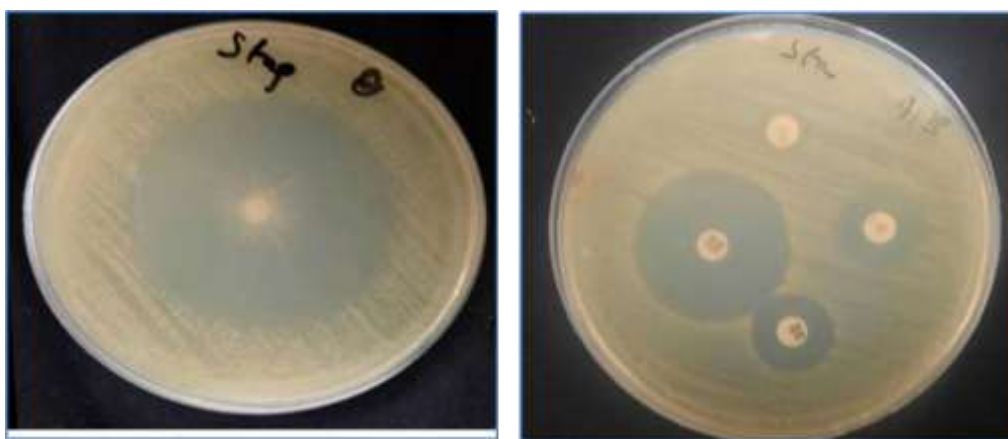
	Oregano	Rosemary	Sage	Thyme	CN	AX	OX	TC
<i>L. innocua</i>	Extr S	R	R	Extr S	Extr S	Extr S	R	Extr S
<i>B. cereus</i>	Extr S	S	R	Extr S	Extr S	Extr S	S	S
<i>S. aureus</i>	Extr S	S	Very S	Extr S	Extr S	S	S	R
<i>E. coli</i>	Extr S	R	Extr S	Extr S	Extr S	Very S	R	S
<i>P. aeruginosa</i>	Extr S	S	R	Extr S	Extr S	R	R	Extr S
<i>C. albicans</i>	Extr S	R	Very S	Extr S	Extr S	R	R	Extr S
Extr S : Extremely sensitive Very S : Very sensitive S : Sensitive R : Resistant								

### 3.2.2. Micro atmosphere method

The antimicrobial activity of the essential oils tested was evaluated by the micro atmosphere method by determining the sensitivity of the different microbial species. The interpretation of the results obtained is done using a scale for estimating antimicrobial activity reported by Ponce *et al.* (2003). Table 3 displays the findings of the assessment of the various EOs' antibacterial activity against the various microbiological strains examined. All investigated microbial strains were susceptible to the potent antibacterial activity of oregano and thyme essential oils with DZI varied from  $12.0 \pm 1.0$  to  $47.5 \pm 2.5$  mm. Sage essential oil, on the other hand, had no antibacterial action against any of the studied microbiological strains with DZI ranging from  $6.0 \pm 0.0$  to  $8.0 \pm 0.0$  mm. With the exception of *L. innocua* and *E. coli*, rosemary essential oil exhibited modest antibacterial activity, but no antimicrobial activity against any of the rest tested strains (Figure 4). While all studied strains were resistant to sage and rosemary essential oils, they were very susceptible to oregano and thyme EOs. But *L. innocua* was more susceptible to the effects of rosemary essential oil. However, *E. coli* was more susceptible to the effects of sage essential oil. Various factors influence the antimicrobial activity of EOs, such as microbial cultures, geographical origin, the part of the plant from which the EOs are derived, the portions used, harvest season, climate, vegetative stage, drying conditions and/or conservation circumstances, and the extraction methodology, which is influenced by several elements such Particle size, solvent concentration and choice, ratio, exhaustion, temperature, pressure, time, and/or extraction method (conventional distillation, Clevenger, Soxhlet) (Mutlu-Ingok, 2020).

**Table 3:** Sensitivity of microbial strains to the EOs tested (Micro-atmospheres)

	Oregano	Rosemary	Sage	Thyme
<i>L. innocua</i>	S	S	R	S
<i>B. cereus</i>	Extr S	R	R	Extr S
<i>S. aureus</i>	Extr S	R	R	Extr S
<i>E. coli</i>	Extr S	S	R	Extr S
<i>P. aeruginosa</i>	Extr S	S	R	Extr S
<i>C. albicans</i>	Extr S	R	R	Extr S
Extr S : Extremely sensitive Very S : Very sensitive S : Sensitive R : Resistant				



**Figure 4:** Sensitivity of microbial strains to the EOs tested for the micro atmosphere and disk diffusion methods

### 3.2.3. MIC determination

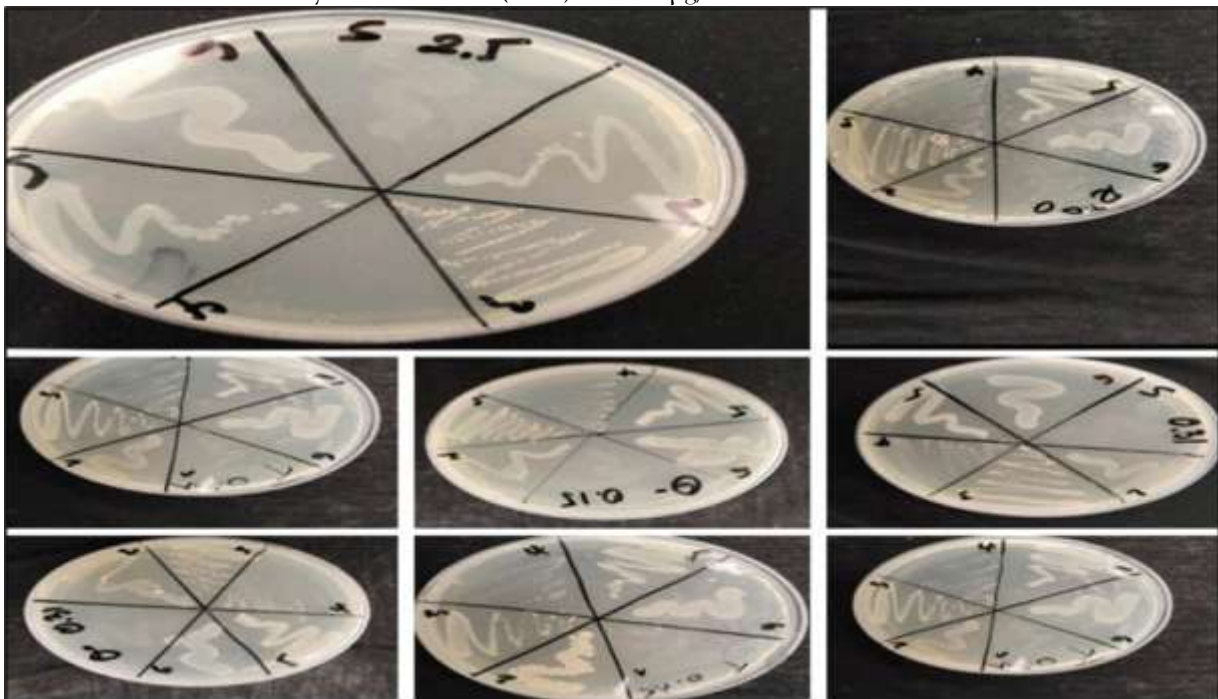
According to Belanger et al. (2021), the minimum inhibitory concentration (MIC) of the essential oils under test is the lowest concentration of an antimicrobial agent needed to prevent a particular microorganism's apparent growth. Table 4 shows the MIC findings for the various EOs examined on the various microbiological strains examined. It is evident from the data in Table 4 that strains of *S. aureus*, *E. coli*, and *L. innocua* can be inhibited from growing at a thyme EO concentration of 0.125 %. However, strains of *Candida albicans*, *B. cereus*, and *P. aeruginosa* can be inhibited from growing at a greater dosage of 0.25% thyme EO. Table 3 shows that an oregano EO concentration of 0.25% allows the inhibition of the growth of all the microbial strains tested. Table 4 shows that at a 0.5% sage EO concentration, neither the *L. innocua* nor the *E. coli* strains exhibit any growth. However, in order to inhibit the *S. aureus* strain from growing, 1% of this EO must be provide. However, a concentration of this EO higher than 1% is necessary to suppress the growth of the strains of *Candida albicans*, *B. cereus*, and *P. aeruginosa*.

**Table 4:** The results of the MIC of essential oils

Concentration (µg/mL)		1	0.5	0,25	0,125	0,062	0,031	0,015	0,0075	0
<i>L. innocua</i>	Thyme	+	+	+	+	-	-	-	-	-
	Oregano	+	+	+	-	-	-	-	-	-
	Sage	+	+	-	-	-	-	-	-	-
	Rosemary	+	-	-	-	-	-	-	-	-
<i>B. cereus</i>	Thyme	+	+	+	-	-	-	-	-	-
	Oregano	+	+	+	-	-	-	-	-	-
	Sage	-	-	-	-	-	-	-	-	-
	Rosemary	+	-	-	-	-	-	-	-	-
<i>S. aureus</i>	Thyme	+	+	+	+	-	-	-	-	-
	Oregano	+	+	+	-	-	-	-	-	-
	Sage	+	-	-	-	-	-	-	-	-
	Rosemary	+	-	-	-	-	-	-	-	-
<i>E. coli</i>	Thyme	+	+	+	+	-	-	-	-	-
	Oregano	+	+	+	-	-	-	-	-	-
	Sage	+	+	-	-	-	-	-	-	-
	Rosemary	+	+	-	-	-	-	-	-	-
<i>P. aeruginosa</i>	Thyme	+	+	+	-	-	-	-	-	-
	Oregano	+	+	+	-	-	-	-	-	-
	Sage	-	-	-	-	-	-	-	-	-
	Rosemary	+	-	-	-	-	-	-	-	-
<i>C. albicans</i>	Thyme	+	+	+	-	-	-	-	-	-

	Oregano	+	+	+	-	ND	-	-	-	-
	Sage	-	-	-	-	-	-	-	-	-
	Rosemary	+	-	-	-	-	-	-	-	-
ND: none detected										

According to the findings in Table 4, all microbiological strains were inhibited at a 1% concentration of rosemary essential oil, except for *E. coli*. However, the growth of *E. coli* can be inhibited by this EO at a lower dose of 0.5%. Additionally, the microbial strains *C. albicans*, *B. cereus* and *P. aeruginosa* exhibit the same sensitivity to the examined EOs (Figure 5). The latter have MICs of 0.25% for thyme and oregano EOs, a MIC of 1% for rosemary EO, and a MIC greater than 1% for sage EO against the tested strains. The microbial strains show sensitivity to the tested EOs. The latter have a similar MIC of 0.25% for oregano EO and a lower MIC of 0.125% for thyme EO. While sage EO shows identical MICs of 0.5% for *E. coli* and *L. innocua* and a MIC of 1% for *S. aureus*. However, rosemary EO has a MIC of 0.5% for *E. coli* and a MIC of 1% for *L. innocua* and *S. aureus*. Antimicrobial activity has been studied by several authors. According to Martins (2020), *B. cereus* growth is inhibited at a MIC of 125 µg/mL of thyme EO, while *E. coli* and *S. aureus* growth is inhibited at a MIC of 500 µg/mL. According to Bouras *et al.* (2019), oregano essential oil (EO) may inhibit *P. aeruginosa* from growing at a MIC of 200 µg/mL and *E. coli* and *S. aureus* from growing at a MIC of 100 µg/mL. According to Stojiljkovic *et al.* (2018), isolates of *E. coli*, *P. aeruginosa*, *B. cereus*, and *S. aureus* were sensitive to rosemary EO at MICs of 200, 480, 1160, and 1790 µg/mL. When thyme EO was tested against the same bacteria, Boulaghmen *et al.* (2018) found that *E. coli*, *P. aeruginosa*, and *Candida albicans* had MICs of 0.125, 0.25, and 0.03%, respectively, whereas *B. cereus* and *S. aureus* had MICs of 0.06%. The growth of *P. aeruginosa* and *E. coli* was demonstrated to be inhibited by sage or rosemary essential oils at a MIC of 25 µg/mL. The antibacterial activity of these essential oils against a few studied pathogens, especially *P. aeruginosa* and *E. coli*, was examined by Kačaniová *et al.* (2017). However, in order to prevent *E. coli* from developing, rosemary essential oil must have a minimum inhibitory concentration (MIC) of 12.5 µg/mL.



**Figure 5:** MIC of essential oils

According to Pesavento *et al.* (2015), with MICs of 0.25, 0.5, 10.0, and 12.5 µg/mL, respectively, the essential oils of thyme, oregano, rosemary, and sage inhibited the growth of *S. aureus*. However, for thyme, oregano, rosemary, and sage essential oils, *L. monocytogenes* was susceptible to MICs of 0.25, 0.0625, 5.0, and 60.0 µg/mL, respectively. Bozin (2007) demonstrated that the growth of *Candida albicans* was inhibited by rosemary and sage essential oils (EOs) with MICs of 30 and 200 µg/mL, respectively.

Generally, EOs' antimicrobial properties are determined by their chemical composition, the type and concentration of the majority of their constituents, the structure and composition of their functional groups, and the probability of a synergistic interaction between them (Andrade *et al.*, 2014). Additionally, the kind of microbial strains evaluated, and the selection of appropriate media may be impacted by the activity evaluation method (Mutlu-Ingok *et al.*, 2019).

## CONCLUSION

The present study demonstrates the significant antimicrobial and antioxidant potential of four essential oils from *Lamiaceae* species: oregano, thyme, rosemary, and sage. Among these, oregano and thyme oils exhibited the highest efficacy against all tested microbial strains, as well as remarkable antioxidant capacities, making them promising candidates for natural applications. In contrast, sage and rosemary oils showed more selective activities. Therefore, this study supports the use of *Lamiaceae* essential oils in food preservation and shelf-life extension by demonstrating that they can further enhance distinctive colors and flavors, thereby substituting synthetic antioxidants. More investigation is required to assess their stability, toxicity, and modes of action in practical. This work underscores the value of natural bioresources in addressing current challenges in health and sustainability.

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*Compliance with Ethical Standards:* This article does not contain any studies involving human or animal subjects.

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**Figure captions:**

Figure 1: Percentage of DPPH inhibition as a function of the concentrations of EOs, BHT and AA.

Figure 2: Percentage of FRAP inhibition as a function of the concentrations of EOs, BHT and AA.

Figure 3: Percentage of TAC inhibition as a function of the concentrations of EOs, BHT and AA.

Figure 4: Sensitivity of microbial strains to the EOs tested for the micro atmosphere and disk diffusion methods

Figure 5: CMI of essential oils