

Citation: Egypt. Acad. J. Biolog. Sci. (G. Microbiolog) Vol.17 (1) pp.135- 149 (2025) DOI: 10.21608/EAJBSG.2025.428543 Egypt. Acad. J. Biolog. Sci., 17(1):135-149 (2025)



Egyptian Academic Journal of Biological Sciences G. Microbiology

> ISSN: 2090-0872 https://eajbsg.journals.ekb.eg/



Antioxidant, Antibacterial Comparative Study between Artemisia monosperma L. and artichoke (Cynara scolymusL.).

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#### **ARTICLE INFO**

Article History Received:12/4/2024 Accepted:19/5//2025 Available:23/5/2025

*Keywords*: Antimicrobial, antioxidant, Artemisia, artichoke, Grampositive, Gramnegative.

#### ABSTRACT

Background: The study focuses on exploring the antioxidant and antimicrobial potentials of Egyptian Artemisia monosperma L. and artichoke (Cynara scolymus L.). These plants are known for their bioactive compounds, yet their comparative phytochemical composition and biological activities remain underexplored. The aim is to evaluate their antioxidant capabilities and antibacterial effectiveness to identify their potential as natural therapeutic agents. Materials and Methods: Methanolic extracts were prepared by airdrying, powdering the plant materials, and extracting bioactive components using methanol. Phytochemical analysis quantified the levels of phenolics, flavonoids, and tannins. Antioxidant activity was determined through the DPPH assay, measuring the IC50 values to reflect antioxidant capacity. Antimicrobial activity was assessed through the agar well diffusion method and the determination of Minimum Inhibitory Concentration (MIC) against selected bacterial strains. Results: Artichoke exhibited higher phytochemical content, including phenolics, flavonoids, and tannins, compared to Artemisia. Antioxidant analysis revealed that artichoke had a stronger scavenging effect with a lower IC50 value (0.134 mg/mL) compared to Artemisia (0.202 mg/mL), though both were less effective than ascorbic acid. Antimicrobial tests showed that artichoke was more effective against Gram-negative bacteria and Staphylococcus epidermidis than Artemisia. MIC results for both extracts were 0.454 mg/mL against Escherichia coli and S. epidermidis, indicating moderate antibacterial activity. Conclusion: The findings indicate that artichoke possesses superior antioxidants and antimicrobial properties compared to Artemisia. While Artemisia exhibited moderate activity, artichoke demonstrated significant bioactive potential, making it a promising candidate for further research and potential therapeutic applications. Both plants warrant further exploration to maximize their pharmacological benefits.

#### **INTRODUCTION**

Today, the tradition and examination of the pharmacological and biological components of plants has markedly increased due to their minimal side effects compared to synthetic drugs (Kim *et al.*, 2015). Many active compounds in plants have been used in traditional medicines, with 60% of people worldwide relying on herbal medicine for various health problems (Mostafa *et al.*, 2018).

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Artemisia monosperma L., a member of the Asteraceae family, is referred to in Chinese herbal medicine as wormwood or sweet annie. It has been used for diseases such as malaria and fever due to its active components like endoperoxide sesquiterpene, artemisinin, essential oils, and lactone (Bora & Sharma, 2011). Many Artemisia species are cultivated or grow widely for use in herbal medicine, often as particularly formulation, tea a in Mediterranean regions (Vouillamoz et al., 2015). Previous studies have highlighted various biological activities of Artemisia including antimicrobial. leaves. antibacterial, antifungal, antimalarial, antiinflammatory, antiallergic, and antitumor properties (Cavar et al., 2012; Mohammed et al., 2022).

Artichoke (Cynara scolymus L.) is considered a popular functional food containing numerous active ingredients utilized in nutraceutical and medical applications al., (Shallan et 2020). Artichoke is primarily produced in countries such as Italy (377,000 tons annually), Spain (224,000 tons), and Egypt (180,000 tons) (FAOSTAT, 2019). It contains a wealth of polyphenols, including hydroxycinnamic acid, phenolic acid, and flavonoids, with various medical applications for all plant parts (Abu-Reidah et al., 2013; Blanco et al., 2018; Dabbou et al., 2016; Durazzo et al., 2013; Jiménez-Moreno et al., 2019). Among its polyphenol groups, cynarin, caffeic acid, and chlorogenic acid are prominent. Other components present in artichoke heads and stems include apigenin, cyanidin caffeoyl glucoside, and luteolin (Rocchetti et al., 2020; Lattanzio et al., 2009; Petropoulos et al., 2018). The aqueous extract of artichoke contains phenolic acids and flavones (Elshamy et al., 2020). Artichokes from Mediterranean regions are noted for their biological activities such as anticancer, antimicrobial, antioxidant, and antifungal effects, with their high caffeic acid content contributing to their antioxidant properties (Sokkar *et al.*, 2020; Rejeb *et al.*, 2020). The aim of this study to compare between the antimicrobial power of *Artemisia monosperma*, *and artichoke* methanolic extracts which is collected from Egypt and explaining by them difference in antioxidants power.

#### MATERIALS AND METHODS Methanolic Extract Preparation for the 3 Tested Samples:

Methanolic extracts of Cynara scolymus L. (artichoke) and Artemisia monosperma L. are prepared through a similar method. The plant materials are collected, air-dried, and ground into fine powder. For extraction, the powdered material is mixed with methanol in a ratio of 1:10 (weight to volume) and stirred continuously for 24-48 hours at room temperature to ensure effective extraction of bioactive compounds. The mixture is filtered using Whatman filter paper or a similar filtration medium to separate the liquid extract from plant residue. Methanol is evaporated under reduced pressure using a rotary evaporator, resulting in crude methanolic extract. The extracts are stored in a cool, dark place for further analysis or applications (Alternemy et al., 2023; Saleh et al., 2024).

### **Reagents and Instruments:**

The reagents utilized included the Folin-Ciocalteu reagent (analytical grade, Fluka, Biochemical Inc., Bucharest, Romania), gallic acid (≥98%, Biomedical Inc., Orange City, FL, USA), 1,1-diphenyl-2-picrylhydrazyl (DPPH•) (>97%). aluminum chloride (anhydrous,  $\geq 99\%$ ), sodium hydroxide (pellets, ≥99%), sodium nitrite (>99%), catechin hydrate (>98%), vanillin (≥99%), hydrochloric acid (37%), and ascorbic acid ( $\geq$ 99%), all sourced from Sigma-Aldrich (St. Louis, MO, USA). Sodium carbonate (analytical grade) and tannic acid (≥98%) were obtained from El-Nasr Pharmaceutical Chemicals (Cairo, Egypt). For the extraction process, a horizontal water bath shaker (Memmert WB14, Schwabach, Germany) was used, while phytochemical analyses and antioxidant activity measurements were performed with a spectrophotometer (Spekol 11, Analytic Jena AG, Jena, Germany) and a UV lamp (Vilber Lourmat-6.LC, VILBER Smart Imaging, Marne-la-Vallée, France).

# Antioxidant Activity Assessment Using DPPH Assay:

The evaluation of antioxidant properties of Artemisia monosperma L. and artichoke (Cynara scolymus L.) was performed using the DPPH• method, a widely utilized colorimetric approach, with ascorbic acid as the reference standard. This analysis adhered to previously established protocols (Alanazi et al., 2025). dilutions were prepared Serial bv combining the samples with equivalent volumes of methanol. A DPPH• solution (concentration: 0.135 mM) was then added to these dilutions in equal volumes. The mixtures were left undisturbed and protected from light for 30 minutes at room temperature. Absorbance measurements were subsequently taken at 517 nm. To determine the percentage of DPPH• remaining, calculations were performed utilizing the equation provided in the study (Eq. (1):

% DPPH remaining =  $[DPPH]_{T/}$ [DPPH]<sub>T=0</sub> x 100 Eq. (1)

percentage of DPPH• The remaining was graphed against the sample concentration (measured in mg/mL) to exponential curve and construct an determine the effective concentration. known as "IC50." The IC50 represents the quantity of antioxidant compounds required to reduce the initial concentration of DPPH• solution by 50%. A lower IC50 value reflects a higher antioxidant capacity of the tested sample, demonstrating an inverse correlation between IC50 and antioxidant efficacy (Parejo et al., 2000).

### **Phytochemical Analysis:**

Folin-Ciocalteu Assay: The phenolic content in the samples was measured using the Folin-Ciocalteu method, as described in (Sánchez-Rangel *et al.*, 2013). In this

process, 100 µL of the sample was placed into a cuvette, followed by the addition of 5 mL of Folin-Ciocalteu reagent. The reagent was prepared by diluting 1 mL of the original solution with 9 mL of distilled water. The mixture was thoroughly mixed and left to incubate for 5 minutes. Subsequently, 4 mL of a 7.5% sodium carbonate solution was added, and the mixture was again stirred. The total volume was then adjusted to 10 mL with distilled water. The solution was kept in the dark at 40°C for 30 minutes, allowing for the development of a blue color. The absorbance was recorded at 765 nm using a spectrophotometer.

To quantify the phenolic content, a standard curve for gallic acid was prepared within a concentration range of 0– 100 mg/L. The absorbance readings from the standards were plotted against their respective concentrations, forming a linear regression equation. This equation (y = 0.0062x, r<sup>2</sup> = 0.987) was used to calculate the phenolic content in the samples. The results were expressed as milligrams of gallic acid equivalents per gram of dry weight (mg GAE/g DW), based on the interpolated sample absorbance values. (y = 0.0062x, r<sup>2</sup> = 0.987).

## Aluminum Chloride Colorimetric Assay:

The flavonoid content of the samples was assessed through the aluminum chloride colorimetric method, as outlined in (Zhishen et al., 1999). Initially, 100 µL of the sample was transferred into a cuvette. To this, 4 mL of distilled water was added, followed by 0.3 mL of a 5% sodium nitrite solution. The mixture was thoroughly combined and allowed to sit for 5 minutes. Subsequently, 0.3 mL of a 10% aluminum chloride solution was mixed thoroughly, introduced, and incubated for 6 minutes. Then, 2 mL of 1 M sodium hydroxide solution was added, followed by thorough mixing, and the mixture was left to stand for 15 minutes at room temperature. The final volume was adjusted to 10 mL using distilled water.

The absorbance of the resulting orange solution was measured at 510 nm with a spectrophotometer. To determine the flavonoid content, a standard curve was created using quercetin concentrations ranging from 0–100 mg/L. The absorbance readings of the standards were plotted against their respective concentrations, forming a linear regression equation. The flavonoid content in the samples was calculated by interpolating their absorbance values into the standard curve equation and expressed as milligrams of quercetin equivalents per gram of dry weight (mg QE/g DW) (y = 0.0028 x, r<sup>2</sup>= 0.988).

## Vanillin-Hydrochloride Assay:

The tannin content in the samples was analyzed through a modified vanillinhydrochloride method, as described by Aberoumand (2009). To perform the assay, 5 mL of freshly prepared vanillinhydrochloride reagent was used. The reagent was made by combining equal parts of 30% hydrochloric acid and methanol with a 4% vanillin solution prepared in methanol. A volume of 1 mL of the sample was added to the reagent and incubated for 20 minutes. After incubation. the absorbance of the orange-colored solution recorded at 510 nm using a was spectrophotometer.

A standard curve based on tannic acid was constructed to convert the absorbance readings into tannic acid equivalents (TAE). The tannin content of the samples was expressed in grams of TAE per 100 grams of dry extract.

### Antibacterial Assessment:

### **Agar Well Diffusion Method:**

To evaluate antimicrobial activity, the agar plate surface is first inoculated by evenly spreading a specified volume of the microbial inoculum across the agar surface. Following this, a sterile cork borer or tip is used to aseptically create a 9 mm diameter well on the plate. A sample volume of 100  $\mu$ L at the desired concentration is then carefully introduced into the well. The plates are subsequently incubated under appropriate conditions

tailored to the test microorganism. During incubation, the antimicrobial agent diffuses into the agar medium, leading to inhibition of the growth of the microbial strain being tested (Elattar *et al.*, 2023).

### Determination of Minimum Inhibitory Concentration (MIC):

To evaluate the MIC, serial dilutions of the sample were prepared in concentrations ranging from 0.057 to 3.63 mg/mL in nutrient broth medium. A control containing only inoculated broth was included and incubated for 24 hours at 37°C. The MIC endpoint was identified as the lowest sample concentration at which no visible microbial growth was observed in the tubes. The turbidity of the tubes was visually examined both before and after the incubation period to confirm the MIC value. Additionally, optical density (OD) measurements were taken at 600 nm for further validation of the results (Parvekar et al., 2020).

## **Statistical Analysis:**

All experimental studies were conducted in triplicate, with the data analyzed through one-way ANOVA to determine mean  $\pm$  standard deviation (M $\pm$ SD). A p-value of less than 0.05 was considered statistically significant.

#### **RESULTS AND DISCUSSION** Phytochemical Analysis:

The phytochemical profile of the recovered samples was determined by quantifying their total phenol, flavonoid, and tannin content (Table 1). Artichoke (Cynara scolymus L.) had a relatively high phenolic composition of 86.14±0.02 mg gallic acid/g of the dry extract,  $14.73\pm0.15$ catechin/g of flavonoids, mg and 17.61±0.05 mg tannic acid/g of tannins. Artemisia monosperma L., however, had a relatively lower phenolic composition  $(49.76\pm0.19 \text{ mg/g})$ , the same levels of flavonoids  $(14.68 \pm 0.08)$ mg/g), and somewhat lower tannin levels  $(14.80\pm0.02)$ mg/g) compared to artichoke. These findings were supported by the results of previous studies conducted by Awad et al. (2020) and De Falco et al. (2015), which reported the high phenolic compound, flavonoid, and tannin content in artichokes, contributing to their powerful antioxidant and therapeutic properties. Similarly, a study performed by Elbalola (2020) highlighted that Artemisia monosperma demonstrated notable, though comparatively lower, levels of phenolics and tannins, alongside its bioactive potential.

**Table 1.** The results of the phytochemical analysis of the investigated extracted samples.

Samples	Phenolics Content <sup>[a]</sup>	Flavonoids Content <sup>[b]</sup>	Tannins Content <sup>[c]</sup>
S1	86.14±0.02	14.73±0.15	17.61±0.05
S2	49.76±0.19	$14.68 \pm 0.08$	$14.80 \pm 0.02$

<sup>[a]</sup> Phenolic Content "mg gallic acid/1 gm dry extract"

<sup>[b]</sup> Flavonoid Content "mg catechin acid/1 gm dry extract"

<sup>[c]</sup> Tannins Contents "mg tannic acid acid/1 gm dry extract"

S1: artichoke (Cynara scolymusL.), S2: Artemisia monosperma L.

#### **DPPH Antioxidant Activity:**

The DPPH reactive radical scavenging assay functioned to evaluate the antioxidant activities of each tested sample. Figures 1 and 2, displays the percentage of DPPH radical remaining at different concentrations together with their scavenging activities. Analysis determined the IC50 values by measuring how much sample quantity was needed to eliminate 50% of the DPPH radicals. The DPPH scavenging activity of sample artichoke scolymusL.) (Cynara showed direct proportionality to its concentration, leading to an increase from 35.04% to 84.54% within the tested range from 0.057 mg/mL to 0.454 mg/mL. The measurement of IC50 for artichoke revealed a value of 0.134 which points mg/mL. to moderate antioxidant properties. The antioxidant capacity of Artemisia monosperma L was found to be lower than what was observed in artichoke. The Artemisia extract reached 87.94% scavenging activity at 0.89 mg/mL while artichoke attained 84.54% scavenging activity at 0.454 mg/mL.

The scavenging activity of 72.34% from Artemisia was slightly inferior to the 84.54% activity of artichoke at their shared concentration point of 0.445 mg/mL. At equivalent concentrations, artichoke exhibited superior antioxidant ability with an IC50 value of 0.134 mg/mL,

which proved greater than the corresponding value of 0.202 mg/mL for Artemisia.

The researchers used ascorbic acid as their reference antioxidant for comparative purposes. These results the samples indicate that while all dose-dependent possessed antioxidant activities, the sample (artichoke) possessed enhanced activity compared to the sample (Artemisia), but less compared to the reference antioxidant, ascorbic acid.

The results of our study on the powerful antioxidant capacity of artichoke (Cynara scolymus) are consistent with the findings reported in the literature by Salekzamani et al. (2019).which highlighted the potent antioxidant properties of artichoke, attributed to its high phenolic content that enhances oxidative stress defense mechanisms. In contrast, research on Artemisia species demonstrated notable antioxidant activity across various plant parts, with compounds such as artemisinin playing a significant role (Lee et al., 2015). Furthermore, a study conducted by Abid and Abachi (2023), which compared Artemisia to other plants, emphasized its bioactive potential. although its antioxidant capacity was found to be slightly lower than that of artichoke in certain assays.

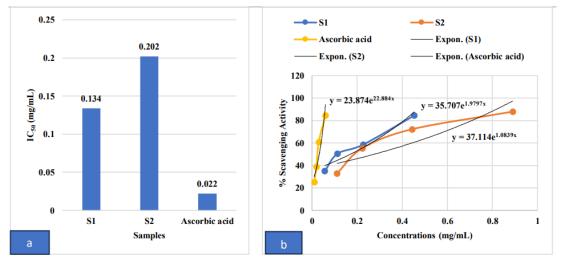


Fig. 1. The antioxidant results by DPPH assay. (a) presented the  $IC_{50}$  values in comparison to ascorbic acid. (b) presented the graph plotted sample concentration versus the % scavenging activity.

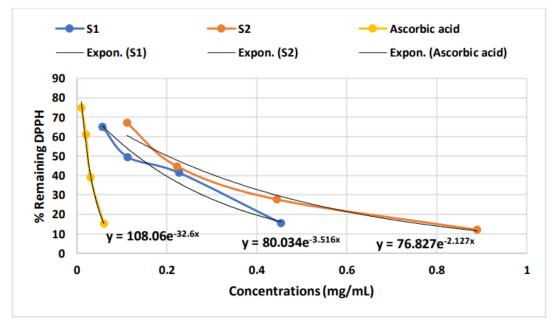


Fig. 2. The relationship between sample concentration (mg/mL) versus % Remaining DPPH.

#### **Antibacterial Activity:**

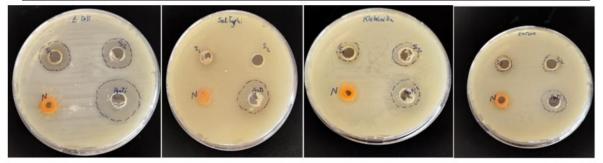
The agar well diffusion method determined antibacterial properties of the tested samples, which were tested against Gram-positive and Gram-negative pathogenic bacteria. The inhibitory region measurements (expressed in millimeter units) appeared in Table 2 and Figure 3. The antibacterial potential of sample artichoke proved most effective against the entire Gram-negative bacteria group. Tests revealed that the product (*artichoke*)

demonstrated the ability to halt the growth of Escherichia coli (ATCC 10536) by  $18.17\pm0.1$  mm in diameter, along with Klebsiella pneumoniae by  $14.07\pm0.20$  mm and Enterobacter cloacae by  $10.99\pm0.21$ mm, yet displayed no inhibition (-ve) on Salmonella typhimurium. The antibacterial activities of Sample *Artemisia* included S. typhimurium inhibition together with inhibitory effects against E. coli and K. pneumonia, and E. cloacae. Tests revealed Gram-positive bactericidal action specifically from artichoke and Artemisia. The antibacterial effect of artichoke was stronger as it blocked Staphylococcus epidermidis growth by 14.04±0.22 mm, but Artemisia only produced an 11.17±0.15 mm inhibition zone for the same organism. The two samples showed no inhibitory action against the bacterial strains Bacillus subtilis, Bacillus cereus, and Staphylococcus aureus. The powerful antibacterial properties of azithromycin served as a reference against all tested strains, whereas the bacteria demonstrated

robust antimicrobial effects. The antibacterial activity of azithromycin created its greatest inhibition zone against Staphylococcus aureus (26.03±0.15 mm), followed by E. coli (24.20±0.20 mm), Bacillus subtilis (23.06±0.21 mm), S. epidermidis (21.10±0.26 mm), and S. typhimurium ( $20.07\pm0.21$  mm). The results indicate that the antibacterial action of Samples *artichoke* and *Artemisia* manifests against selected Gram-negative bacteria and S. epidermidis.

**Table 2.** Antibacterial activity is expressed as Inhibition zones in mm of the tested samples against pathogenic bacteria.

Microorganisms	Sample 1 S1 (artichoke (Cynara scolymusL.)	Sample 2 S2 (Artemisia monosperma L)	Azithromycin
Gram-negative bacteria			
Escherichia coli (ATCC 10536)	18.17±0.15	17.03±0.16	24.20±0.20
Salmonella typhimurium (ATCC 25566)	-ve	-ve	20.07±0.21
Klebsiella pneumonia (ATCC 10031)	14.07±0.20	14.99±0.21	$19.07 \pm 0.50$
Enterobacter cloacae (DMS 30054)	10.99±0.21	12±0.33	15.17±0.20
Gram-positive bacteria			
Bacillus subtilis (DMS 1088)	-ve	-ve	23.06±0.21
Bacillus cereus (EMCC number 1080)	-ve	-ve	12±0.44
Staphylococcus aureus (ATCC 6538)	-ve	-ve	26.03±0.15
<i>Staphylococcus epidermidis</i> (EMCC number 1353 <sup>t</sup> )	14.04±0.22	11.17±0.15	21.10±0.26

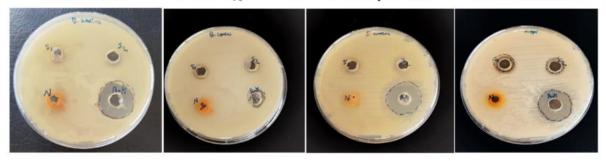


Escherichia coli

Salmonella typhimurium

Klebsiella pneumonia

Enterobacter cloacae



*Bacillus subtilis Bacillus cereus Staphylococcus aureus Staphylococcus epidermidis* **Fig. 3**. The Petri dish images conceding the antibacterial impact of the samples.

Studies have extensively discussed the antibacterial activity of artichoke extracts against Gram-negative bacteria, primarily highlighting phenolic compounds such cynarin as and chlorogenic acids as effective inhibitors of microbial growth. Research by Yildirim et al. (2020) and Abd El-Ghany (2017) demonstrated the significant activity of artichoke extracts against Escherichia coli and Klebsiella pneumoniae, with inhibition zones measuring 18.17±0.15 mm and 14.07±0.20 mm, respectively. Notably, while artichoke extracts show promising against many Gram-negative results bacteria, they exhibit no inhibitory activity typhimurium. Salmonella against consistent with findings from other studies (Gavriil et al., 2021; De Falco et al., 2015; Guerrero-Encinas et al., 2024). This selective activity may be attributed to variations in bacterial cell wall structures or resistance mechanisms.

Regarding Gram-positive bacteria, Abd El-Ghany (2017) reported that artichoke extracts generally show limited inhibition against Bacillus subtilis and Bacillus cereus, but exhibit notable activity Staphylococcus against epidermidis. aligning with findings of a 14.04±0.22 mm inhibition zone. The efficacy of artichoke extracts appears to vary based on extraction methods; for example, ethanolic extracts display greater inhibitory effects against Staphylococcus aureus, while methanolic extracts are less effective (Gaafar & Salama, 2013; Abd El-Ghany, 2017). Optimizing extraction techniques is therefore critical for maximizing antibacterial potential.

Artemisia extracts also demonstrate antibacterial activity against Gram-negative bacteria, with inhibition zones of  $17.03\pm0.16$  mm for Escherichia coli and  $14.99\pm0.21$  mm for Klebsiella pneumoniae, supported by other research (Talib *et al.*, 2020; Mohammed *et al.*, 2022; Einollah *et al.*, 2012) exploring various Artemisia species. These effects are attributed to bioactive compounds such as artemisinin and flavonoids, which disrupt bacterial cell walls. However, Artemisia extracts show limited or no activity against Salmonella typhimurium, consistent with prior studies (Mohammed *et al.*, 2022; Bordean *et al.*, 2023; Ahameethunisa & Hopper, 2010).

For Gram-positive bacteria, Artemisia extracts exhibit an 11.17±0.15 mm inhibition zone against Staphylococcus epidermidis, attributed to the interaction of bioactive compounds with bacterial cell walls. However, limited or no activity is observed against Bacillus subtilis, Bacillus and Staphylococcus cereus, aureus. aligning with previous research (Mohammed et al., 2022; Bordean et al., 2023; Nametov et al., 2023).

In comparing the antibacterial efficacy of artichoke and Artemisia extracts, no significant difference is observed for Gram-negative bacteria. However, artichoke demonstrates greater activity against Gram-positive Staphylococcus epidermidis (EMCC number 1353t) than Artemisia, potentially due to its higher phenolic content and strong scavenging activity.

# Minimum Inhibitory Concentration (MIC):

The MIC of samples *artichoke* and Artemisia was determined against *Staphylococcus* Escherichia coli and epidermidis using the broth dilution method. Serially diluted samples and microbial growth visually observed by turbidity and spectrophotometrically measured through OD<sub>600</sub> measurements after 37 °C at 24 hours' incubation were observed. The MIC was assumed as the least concentration at which there was absence of visible growth and OD<sub>600</sub> was also found at baseline value.

## MIC Values for *Escherichia coli*:

In *artichoke* and *Artemisia*, turbidity first appeared in tube 5, indicating microbial growth at a level of 0.227 mg/mL and below. The MIC was therefore determined to be 0.454 mg/mL for both samples (Tables 3,4 and Fig. 4).

Test tube no.	Concentration (mg/ml)	<b>O.D</b> <sub>600</sub>
1	3.63	0.025
2	1.815	0.015
3	0.908	0.017
4	0.454	0.011
5	0.227	0.887
6	0.113	1.281
7	0.057	1.364

**Table 3.** The measured O.D. 600 for *E. coli* for (S1) artichoke (Cynara scolymusL.)

Table 4. The measured O.D 600 for E.	. <i>coli</i> for sample (S2) <i>Artemisia monosperma L</i>
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Test tube no.	<b>Concentration (mg/ml)</b>	<b>O.D</b> 600
1	3.56	0.026
2	1.78	0.094
3	0.89	0.029
4	0.445	0.034
5	0.223	1.217
6	0.111	1.273
7	0.056	1.275

#### Spectrophotometric Readings Were in Accordance with the Visual Observations:

For *artichoke*, OD<sub>600</sub> was low at 3.63 to 0.454 mg/mL (OD<sub>600</sub> = 0.025 to 0.011) but increased sharply at 0.227 mg/mL (OD<sub>600</sub> = 0.887) and further diluted, confirming the initiation of bacterial growth.

For *Artemisia*, the same trend was observed with OD<sub>600</sub> 0.026 to 0.034 for 3.56 to 0.445 mg/mL and an unexpected peak at 0.223 mg/mL (OD<sub>600</sub> = 1.217), validating 0.445 mg/mL to be the MIC.

# **MIC Results for** *Staphylococcus epidermidis*:

Likewise, for *S. epidermidis*, both *artichoke* and *Artemisia* also showed visible turbidity from test tube 5 onwards, corresponding to concentrations of 0.227 mg/mL and below. The MIC for both samples was therefore 0.454 mg/mL (Tables 5,6 and Fig. 4).

# The OD<sub>600</sub> Readings Supported This Result:

For *artichoke*, the OD was low (0.031 to 0.052) from 3.63 to 0.454 mg/mL but increased at 0.227 mg/mL (OD<sub>600</sub> = 0.578), indicating growth beyond this concentration.For *Artemisia*, the OD<sub>600</sub> was between 0.097 and 0.033 up to 0.445 mg/mL and then jumped abruptly at 0.223 mg/mL (OD<sub>600</sub> = 1.137), confirming the MIC at 0.445 mg/mL.

These results clearly indicate that *artichoke* and *Artemisia* both possess moderate antimicrobial activity, with repeated MIC values of 0.454 mg/mL against *E. coli* and *S. epidermidis*. This supports the findings of the agar well diffusion assay and suggests potential utility for these samples as antibacterial agents, but their activity is limited in comparison to conventional antibiotics.

Test tube no.	Concentration (mg/ml)	<b>O.D</b> <sub>600</sub>
1	3.63	0.031
2	1.815	0.016
3	0.908	0.098
4	0.454	0.052
5	0.227	0.578
6	0.113	0.712
7	0.057	0.766

**Table 5.** The measured O.D 600 for S. epidermidis for sample (S1) artichoke (Cynara scolymusL.)

Table 6. The measured O.D600 for S	epidermidis for sample	ole (S2) Artemisia monosperma L
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Test tube no.	<b>Concentration (mg/ml)</b>	<b>O.D</b> 600
1	3.56	0.097
2	1.78	0.054
3	0.89	0.127
4	0.445	0.033
5	0.223	1.137
6	0.111	1.219
7	0.056	1.207

Both artichoke and Artemisia extracts demonstrated moderate antimicrobial activity against Escherichia coli and Staphylococcus epidermidis, as revealed by the broth dilution method. The minimum inhibitory concentration (MIC) was consistently determined to be 0.454 mg/mL for both samples against both bacterial strains, aligning with findings reported in Tables 4–6. Spectrophotometric readings supported these results. showcasing similar trends in OD600 measurements for both extracts, indicating microbial growth initiation at concentrations below the MIC. Notably, Escherichia coli showed comparable sensitivity to both extracts, with OD<sub>600</sub> readings sharply increasing at 0.227 mg/mL, corroborating bacterial growth beyond the MIC, as highlighted by Abd El-Ghany (2017).

For Staphylococcus epidermidis, while both extracts inhibited bacterial growth up to 0.454 mg/mL, Artemisia exhibited an unexpected peak in OD<sub>600</sub> measurements at 0.223 mg/mL (OD<sub>600</sub> = 1.137), slightly higher than artichoke's corresponding value (OD<sub>600</sub> = 0.578), as also observed in prior studies (Mohammed *et al.*, 2022; Bordean *et al.*, 2023). This suggests slight variability in bacterial response to Artemisia's active compounds, such as artemisinin and flavonoids.

Overall, while both extracts showed consistent MICs, the results underscore their limited efficacy compared to conventional antibiotics, supporting the findings of Yildirim *et al.* (2020) and other related research (Gavriil *et al.*, 2021; Guerrero-Encinas *et al.*, 2024; Nametov *et al.*, 2023).

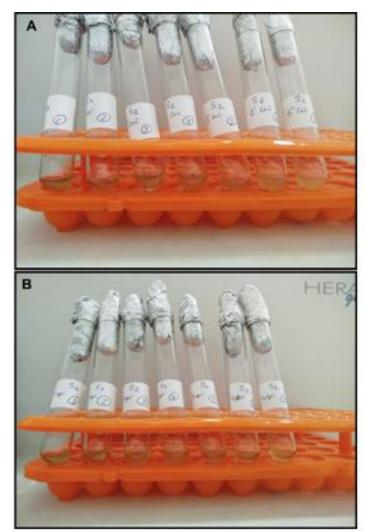


Fig. 4. MIC detection of *artichoke* (S1) and *Artemisia* (S2) (3.56 mg/mL, 1.78mg/mL, 0.89 mg/mL, 0.445 mg/mL, 0.223 mg/mL, 0.111 mg/mL, 0.056 mg/mL) against *E. coli* and *S. epidermidis* showing MIC at 0.445 mg/mL.

#### CONCLUSION

The present study concluded that artichoke (Cynara scolymus L.). exhibited higher levels of phenolics, flavonoids, and tannins, resulting in stronger antioxidant capabilities than Artemisia monosperma L. Antimicrobial analysis revealed artichoke to be more effective against specific bacteria, including Gram-negative strains and Staphylococcus epidermidis, compared While to Artemisia. both samples demonstrated moderate antibacterial activity (MIC of 0.454 mg/mL), their efficacy was limited compared to standard antibiotics. Overall, artichoke showcased superior antioxidant and antimicrobial potential, while Artemisia offered modest yet notable benefits.

#### List of abbreviations

- AlCl<sub>3</sub>: Aluminum Chloride
- **ANOVA**: Analysis of Variance
- **DPPH**•: 2,2-Diphenyl-1picrylhydrazyl (free radical used in antioxidant assays)
- **DW**: Dry Weight
- FAOSTAT: Food and Agriculture Organization Corporate Statistical Database
- GAE: Gallic Acid Equivalent
- **HCl**: Hydrochloric Acid
- IC50: Half-maximal Inhibitory Concentration
- **M±SD**: Mean ± Standard Deviation
- MIC: Minimum Inhibitory Concentration
- Na<sub>2</sub>CO<sub>3</sub>: Sodium Carbonate
- NaOH: Sodium Hydroxide
- NaNO<sub>2</sub>: Sodium Nitrite

- **OD**: Optical Density
- **QE**: Quercetin Equivalent
- **r**<sup>2</sup>: Coefficient of Determination
- **TAE**: Tannic Acid Equivalent
- UV: Ultraviolet

## **DECLARATIONS:**

Ethical Approval: Not applicable.

Authors' Contributions: LAN conceptualized, designed the research protocol, participated in the entire research work and wrote the first and final draft of the manuscript.

**Declaration of Competing Interest:** The author declares no competing interests.

**Data availability Statement:** All data used for the study are available in the manuscript.

Funding: No funding was received.

Acknowledgment: This study was done in the Department of Biology, Faculty of Science, Umm Al-Qura University, Makkah, Saudi Arabia.

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