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Effect of Nanopraticles of *Achillea Millefolium* Oil against Pathogenic Bacteria and Antioxidant Activity

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ABSTRACT: The potential of yarrow (*Achillea millefolium*) as a drug in folk medicine is of interest because of its medicinal applications. The antibacterial, and antioxidant activities and the chemical characteristics of the essential oil nanoparticles were investigated in this study. The silver nanoparticles were studied using the UV-vis spectroscopy, FTIR, GC-MS, and atomic force microscopy. The antibacterial efficacy of the nanoparticles was assessed against different bacteria, such as *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Klebsiella pneumoniae*, employing the well diffusion method. It was employed for its DPPH radical scavenging activity and its antioxidant effects. The production of NPs was established by the color change and the spectral shift observed at 219 nm using UV-vis spectroscopy, which indicated that there was a surface plasmon resonance peak at $\lambda = 219$ nm. The resulting AgNPs exhibited a spherical morphology with an average diameter of approximately 57.80 nm. The analysis of the FTIR spectra of the *A. millefolium* oil nanoparticles revealed functional groups. Certain chemicals in these nanoparticles were identified through GC-MS studies. The antibacterial and antioxidant efficacy of *A. millefolium* essential oil nanoparticles was significant. It was concluded that AgNPs of *A. millefolium* have antibacterial and antioxidant activities.

1. INTRODUCTION

Nanoparticles from Plant and Vegetal Materials The utilization of natural plant-based materials to obtain nanoparticles is extremely attractive, not only because of their therapeutic use but also because of the “green” environment in which the nanoparticles can be produced (Liyana and Jasmine, 2025). As a source of naturally occurring phenolic compounds, yarrow is a medicinal plant with a broad spectrum of bioactivities, including anti-inflammatory, antibacterial, and antioxidant properties (Issa and Mahdi, 2024; Fantasma et al., 2024).

The EOs of *A. millefolium* are composed of bioactive compounds such as terpenoids, flavonoids, and sesquiterpene lactones and are known for their biological activities (Konarska et al., 2023). One of the well-established approaches to improve the effectiveness of bioactive molecules in addition to increasing their bioavailability, solubility, and precise delivery, is to encapsulate them into nanoparticles (Haque et al., 2019). Owing to nanoparticles and their large surface area, the antimicrobial and antioxidant activities of these kinds of natural extracts are increased (Shami, 2025).

Many studies have been conducted to assess the antibacterial activity of nanoparticles produced from plant extracts against harmful microorganisms such as *E. coli*, *S. aureus*, and *P. aeruginosa* (Singh et al., 2020; Birusanti et al., 2025). These microorganisms cause different diseases among humans and have become resistant to standard antibiotics; therefore, alternative treatments have gained the interest. Nanoparticles of *A. millefolium* essential oil might be a potential way to prevent the growth of pathogens through the cell wall lysis of the microorganisms, alteration of cellular metabolism, and induction of oxidative stress (Sadeghi, and Gholamhoseinpoor, 2015; Rathore, et al., 2018).

The antibacterial and antioxidant capacities of *A. millefolium* oil contained in nanoparticles should not be overlooked. A key factor in oxidative stress is the imbalance between free radicals and antioxidant defense, which is linked to many long-term illnesses, including cancer, heart disease, and neurological problems. Antioxidants exhibit excellent scavenging activity, prevent cell damage, and abundance of bioactive components in natural extracts (Gupta and Chauhan, 2021; Hamza and Nabila, 2023).

The aim of this study was to synthesize nanoparticles with AEO *A. millefolium*, which can be used as a possible technique to produce alternative antibacterial or oxidative agents. These nanocomposites may have potential as substitutes for natural drugs and preservatives in the pharmaceutical and food industries.

2. MATERIALS AND METHODS

2.1 Plant Materials and Extraction of Essential Oils

The plants, which were given to the University of Baghdad's Department of Biology, Faculty of Sciences, with voucher specimen number 1317, were harvested in September 2024 in Erbil (44° 30' 01" E to 44° 34' 30" E). The plants were dried and milled to powder into mill and stored in hermetically closed containers until use.

2.2 Extraction of Essential Oils

The EO was obtained from dried plant material via hydrodistillation in a Clevenger apparatus for 3-4 h. Anhydrous sodium sulfate was efficiently employed to separate and dehydrate the oil stored in amber bottles at 4°C, before being maintained at +4°C in a dark, air-protected environment (Shakeri et al., 2016).

2.3 Synthesis of silver particles (AgNPs)

Preparation of silver nanoparticles through a decrease in silver nitrate using an aqueous extract. In this process, 10 mL of isolated essential oil was combined with 90 mL of a 1 mM silver nitrate (AgNO_3) mixture. The mixture was stirred at room temperature during 4-6 hours. The solution was kept at ambient temperature for 3-4 h, during which its color changed from pale yellow to deep brown, indicating the production of silver nanoparticles (Abd Qasim and Yaaqoob, 2023).

2.4 Evaluation of Silver Nanoparticles

The nanoparticles were analyzed using several techniques. The UV-Vis optical spectrum of the Ag colloid was acquired via a UV-Vis spectrometer (SP8001, Metertech). The UV-Vis absorbance spectra of the material were acquired via a Perkin Elmer spectrophotometer (Elbossaty, 2017). The functional groups present in the plant extract of the AgNPs were confirmed using the FTIR (8400 Shimadzu), and measured from 400 to 4000 cm^{-1} at a resolution of 4 cm^{-1} . The preparations were made by placing a drop of the sample on a glass slide, via standard procedures. This substance was studied in other samples. The plant oil nanoparticles were examined via GC-MS (Agilent Technologies 7820A, USA). A mass spectrometer was used for electron ionization detection at an accelerating voltage of 400 V and mass range from m/z 50-600 was used in these experiments. The gas chromatographic column (30 m length $\times$ 250 μm inner diameter $\times$ 0.25 μm film thickness) was subjected to a temperature program ranging from 50°C to 300°C at a rate of 1°C per minute. Here, the flow rate of the gas was 1.2 ml/min, and the sample was injected with 1 μL during the analysis of extract samples. The components were determined on the basis of their retention period and mass fragmentation patterns compared with the standard data available in the NIST library found at these sites. The particle morphology and structure were identified, and AFM was employed to manipulate the material at the nanometer level. It carries qualitative and quantitative information on three-dimensional (3D) objects for many physical properties, including dimensionality, morphology, and surface roughness (Chaudhuri et al., 2016).

2.5. Antimicrobial Efficacy

The antibacterial efficacy of *A. millefolium* essential oil nanoparticles was assessed against pathogenic bacteria using an agar well diffusion assay. New Muller-Hinton agar plates were inoculated with fresh bacterial cultures. Wells measuring 6 mm in diameter were created using prepared agar, and 50 μL of nanoparticle suspension at concentrations of 100, 50, 25, 12.5, and 6.25 $\mu\text{g/mL}$ was introduced into each well. The plates were subjected to incubation at 37°C for 24 hours, and the inhibition zones were measured on the basis of the antibacterial activity, which was determined by the diameter observed surrounding the well (IS et al., 2024). For MIC method, Nutrient broth (Difco, Detroit, MI, USA) was used as the medium to culture bacteria. One ml of this broth was added to numbered tubes 1-9. One ml of the stock culture was added to tube 1 and

successively diluted until tube number 7. The last 1 ml of tube 7 was discarded. Tube number 8 was used as a negative control and tube 9 as a positive control (Shami, 2018). MBC values were determined by sub-culturing from the MIC assay tubes onto Muller- Hinton agar (Difco, Detroit, MI, USA) and then determining the dilution at which growth was detected.

2.6. Antioxidant Efficacy

Antioxidant activity of *A. millefolium* essential oil nanoparticles DPPH (2,2-diphenyl-1-picrylhydrazyl).The antioxidant capacity of *A. millefolium* essential oil nanoparticles was assessed via DPPH radical scavenging, as described by Shami with some modification (Shami, 2018). In summary, one ml of 0.1 mM DPPH dissolved in ethanol was combined with different quantities of nanoparticle suspensions. The resulting product was subsequently incubated in the dark for 30 minutes, after which its absorbance at 517 nm using a spectrophotometer. The percentage of the activity of DPPH radical scavenging was determined on the basis of absorbance measurements.

2.7 Statistical Analysis

The experiments were conducted thrice, and the data are presented as the average $\pm$ standard deviation (SD). Statistical examination: The data were analyzed statistically using one-way ANOVA and subsequently compared via Tukey's post hoc test for multiple comparisons. P values less than 0.05 indicated statistical significance.

3. RESULTS AND DISCUSSION

3.1 Characterization of Silver Nanoparticles

The green synthesis of silver nanoparticles (AgNPs) utilizing *A. millefolium* essential oil was studied through several analyses. The change in the color of the reaction mixture from pale yellow to dark brown indicated the formation of nanoparticles, which was the activation of surface plasmon resonance frequencies (Sadeghi and Gholamhoseinpoor, 2015). This study is the optimization of a simple method for the synthesis and the characterization of nanoparticles of plant oil using spectroscopic analysis.

3.2 Ultraviolet-Visible Spectroscopy

The measurements were conducted at 25°C using a Shimadzu Model 5000 method to determine and quantify the synthesis the NPs. The stability of the AgNPs was evaluated via UV-vis spectroscopy, as the resonance band of silver is contingent upon the structure and morphology of the synthesized nanoparticles. The components of the plant extract promote the transformation of silver ions into silver atoms (Shahzadi et al., 2025). The UV-vis spectrum of the AgNO₃ solution with plant extracts is presented in Figure 1. The absorption peak at 219 nm suggests that some chemical compounds capable to reacting with silver ions are still present in the intermediate. The peak observed at 219 nm represents the intrinsic interband electronic transition from the 4d valence band to the 5sp conduction band of silver, which is independent of the SPR effect. For a few more months, biogenic AgNPs were monitored and exhibited the same peak wavelength of surface plasmon resonance as that by UV visible spectroscopy until 1 year.

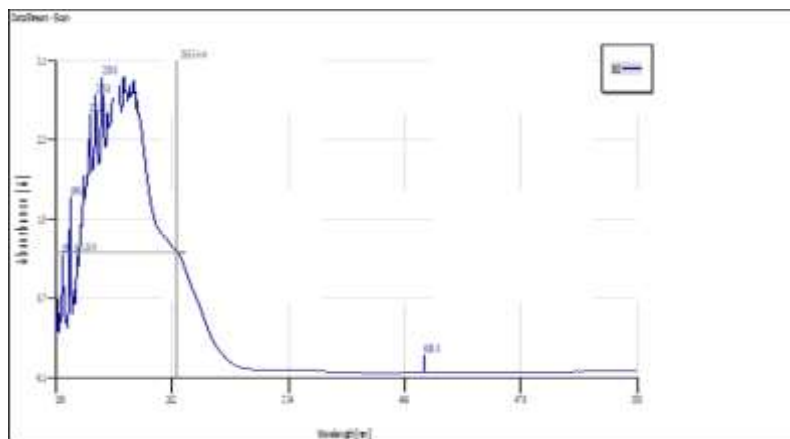


Figure 1.

3.3 FTIR spectroscopy

The FTIR spectra were recorded using a Nicolet 6700 FTIR spectrometer with KBr, which spans from 400 to 4000 cm^{-1} at a resolution of 4 cm^{-1} . FTIR investigation revealed the presence of multiple functional groups, notably hydroxyl and carboxyl groups in the essential oil of *A. millefolium*, which acted as reducing and stabilizing agents in the production of silver nanoparticles. Bioactive chemicals enabled the reduction of Ag^+ to Ag^0 , leading to the creation of nanoparticles, and preventing the aggregation of AgNPs (Tavallaei et al., 2025).

The IR spectra (Figure 2) revealed that strong absorption bands of H stretching occurred at 3474-2590 cm^{-1} for *A. millefolium* essential oil nanoparticles. These C-H stretching vibrations were observed in the wavenumber range of 2882.46–3071.67 cm^{-1} . C=C stretching over reported wavenumbers from 2055.05 to 2363.46 cm^{-1} and C=O stretching from 1733.48-1779.49 cm^{-1} were identified. The C-H bending vibrations peak can be observed at 1303.06 cm^{-1} , and the C-N stretching bands were observed at 1370.06 cm^{-1} and 1044.70 cm^{-1} . Consequently, five principal categories of functions attributed to *A. millefolium* essential oil have been identified (Amato et al., 2025). The major functional major categories of published investigations function of *A. millefolium* essential oils used to prevent parasitic activity in poultry have been investigated.

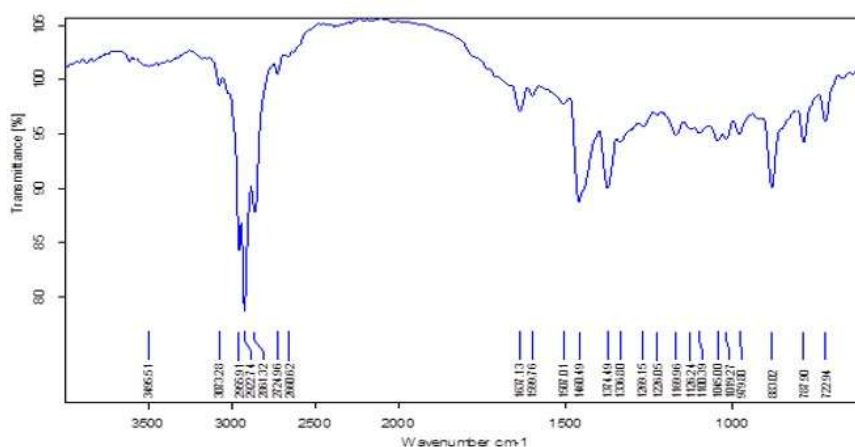


Figure 2.

3.4 GC-MS Analysis

Compounds were also identified by GC-MS, and their mass spectrum fragmentations were compared with those reported in the NIST database library at various percentages of the peak detected (Table 1 and Figure 3). The chief constituents are Alpha-Pinene, 3-Carene, Eucalyptol, Decane, Thujone, Camphor, Endo-Borneol, and 1,3,8-p.Beta-Menthatriene. Bourbonene, Caryophyllene, beta-copaene, gamma-murolene, beta-guaiene, Humulene, beta-cubebene, gamma-murolene, triisopropylchlorosilane, beta-guaiene, camphene and 2,4-nonadienal. Ćimović et al. (2025) performed a GC-MS evaluation of *A. millefolium* L. EB oil nanoparticles along with cyclohexane 1-methylene-4-(1-methyl-ethyl) compounds (44.5%), chamazulene (10.64%), and eucalyptol (8.2%).

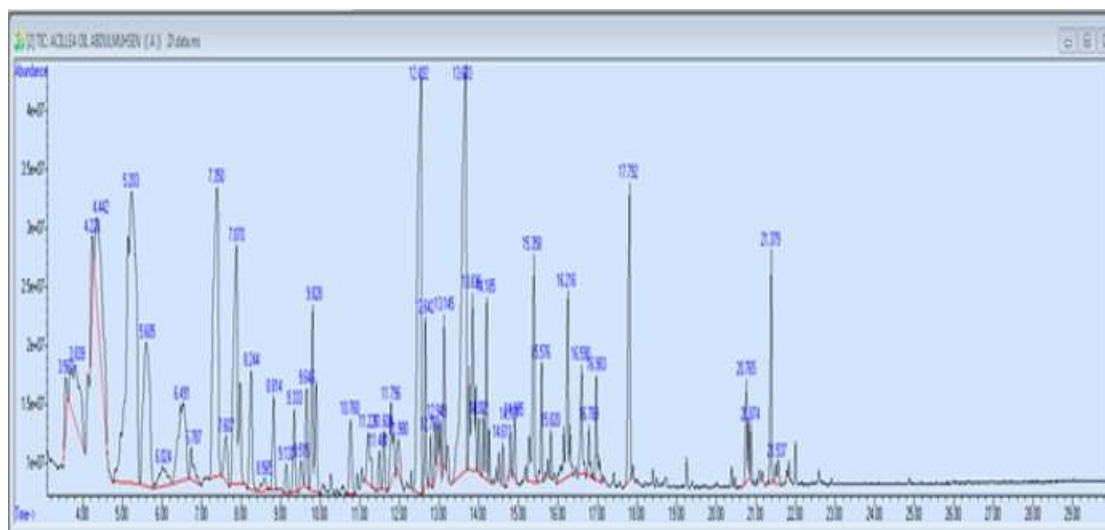


Figure 3.

Table 1. . The GC-MS investigation of *A. millefolium* oil nanoparticles

S. ^a N.	Peak RT ^b (m in)	peak area %	Compound detected*	CAS No. ^c	Molecular Formula ^d	Mol. Wt. ^f g/mol
1	3.563	0.64	Alpha-Pinene	80-56-8	C ₁₀ H ₁₆	136.23
2	3.840	2.68	3-Carene	13466-78-9	C ₁₀ H ₁₆	136.23
3	4.221	0.92	beta.-Pinene	127-91-3	C ₁₀ H ₁₆	136.23
4	4.446	4.15	Bicyclo[3.1.1]heptane	286-34-0	C ₇ H ₁₂	96.17
5	5.199	14.29	Eucalyptol	470-82-6	C ₁₀ H ₁₈ O	154.25
6	5.606	4.76	3-Methyl-2-butenic acid	34876-10-3	C ₁₀ H ₁₄ O ₃	182.22
7	6.021	0.80	2H-pyran	289-66-7	C ₅ H ₆ O	82.10
8	6.488	3.08	Decane	124-18-5	C ₁₀ H ₂₂	142.28
9	6.791	0.50	Thujone	546-80-5	C ₁₀ H ₁₆ O	152.23
10	7.354	7.20	Camphor	76-22-2	C ₁₀ H ₁₆ O	152.23
11	7.605	0.75	Neopentylidenecyclohexane	39546-80-0	C ₁₁ H ₂₀	152.28
12	7.873	5.34	Endo-Borneol	464-43-7	C ₁₀ H ₁₈ O	154.25
13	8.245	1.57	2-Carene	4497-92-1	C ₁₀ H ₁₆	136.23
14	8.566	0.28	Pentane	109-66-0	C ₅ H ₁₂	72.15
15	8.817	1.06	1,3,8-p-Menthatriene	18368-95-1	C ₁₀ H ₁₄	134.22
16	9.137	0.31	Linalyl acetate	115-95-7	C ₁₂ H ₂₀ O ₂	196.29
17	9.336	0.96	cis-Pinen-3-ol	43119-66-0	C ₁₀ H ₁₆ O	152.23

18	9.518	0.36	Decaprenoic acid	459-80-3	C ₁₀ H ₁₆ O ₂	168.23
19	9.648	0.97	Bromoacetyl bromide	598-21-0	C ₂ H ₂ Br ₂ O	201.84
20	9.829	2.76	Isobornyl formate	1200-67-5	C ₁₁ H ₁₈ O ₂	182.26
21	10.764	0.94	1,5-Cyclodecadiene	015423-57-1	C ₁₀ H ₁₆	136.23
22	11.223	0.89	8-dien-2-ol	13544-59-7	C ₁₀ H ₁₆ O	152.23
23	11.483	0.33	beta.-Myrcene	123-35-3	C ₁₀ H ₁₆	136.23
24	11.612	0.56	Copaene	3856-25-5	C ₁₅ H ₂₄	204.35
25	11.794	1.14	beta.-Bourbonene	5208-59-3	C ₁₅ H ₂₄	204.35
26	11.985	0.40	3,5-Heptadienal	099172-18-6	C ₇ H ₁₀ O	110.15
27	12.495	7.75	Caryophyllene	87-44-5	C ₁₅ H ₂₄	204.35
28	12.642	1.10	beta.-copaene	1000374-18-9	C ₁₅ H ₂₄ O	220.35
29	12.781	0.33	beta-Farnesene	28973-97-9	C ₁₅ H ₂₄	204.35
30	12.945	0.96	cis-beta-Farnesene	28973-97-9	C ₁₅ H ₂₄	204.35
31	13.144	1.34	Humulene	6753-98-6	C ₁₅ H ₂₄	204.35
32	13.603	9.37	beta-Cubebene	13744-15-5	C ₁₅ H ₂₄	204.35
33	13.837	2.88	Camphene	79-92-5	C ₁₀ H ₁₆	136.23
34	14.001	0.40	Thujopsene	470-40-6	C ₁₅ H ₂₄	204.35
35	14.183	1.96	Naphthalene	91-20-3	C ₁₀ H ₈	128.17
36	14.607	0.29	Cadala-1(10),3,8-triene	1000140-05-6	C ₁₅ H ₂₂	202.33
37	14.789	0.33	Spiro[4.5]decane	176-63-6	C ₁₀ H ₁₈	138.25
38	14.901	0.39	alpha-Farnesene	502-61-4	C ₁₅ H ₂₄	204.35
39	15.360	2.61	4-Hexadecen-6-yne	74744-51-7	C ₁₆ H ₂₈	220.39
40	15.576	0.90	Longifolene	475-20-7	C ₁₅ H ₂₄	204.35
41	15.819	0.28	Gleenol	1000374-18-1	C ₁₅ H ₂₆ O	222.37

42	16.217	2.45	gamma-Murolene	030021-74-0	C ₁₅ H ₂₄	204.35
43	16.589	1.09	beta.-Guaiene	88-84-6	C ₁₅ H ₂₄	204.35
44	16.788	0.41	gamma-himachalene	1000140-08-0	C ₁₅ H ₂₄	204.35
45	16.979	0.95	Chamazulene	529-05-5	C ₁₄ H ₁₆	184.28
46	17.792	3.11	Triisopropylchlorosilane	13154-24-0	C ₉ H ₂₁ ClSi	192.80
47	20.761	1.14	10-Camphorsulfonyl chloride	21286-54-4	C ₁₀ H ₁₅ ClO ₃ S	250.74
48	20.874	0.28	3-Butylpyridine	539-32-2	C ₉ H ₁₃ N	135.21
49	21.375	1.73	2-4-nonadienal	000079-06-1	C ₁₀ H ₁₆ O	152.23
50	21.540	0.33	2-Propenamide	000079-06-1	C ₆ H ₇ NO ₂	125.13

3.5. Atomic Force Microscopy (AFM)

The shape and appearance of the *A. millefolium* oil nanoparticles were further examined using AFM imaging. Figure 4 shows that the nanoparticles displayed a spherical morphology, characterized by a smooth and uniform surface topography, with an average particle size of 57.8 nm. The homogeneity of the size during production is a crucial factor that affects the durability and efficacy of nanoparticles in biological systems. The height profiles obtained from AFM images show an average particle height of 57.8 nm. The cross-sectional analysis suggests a quasi-spherical morphology on the mica substrate. This finding corresponds with the peak distribution histogram data, indicating a restricted particle dimension range. This confirmed findings that were likewise in agreement. As Bhat et al. (2021). The main produced silver nanoparticles were spherical, measuring between 25 and 50 nm, or appeared as clusters of 100 nm. Githala et al. (2022) examined the dimensions and morphology of nanoparticles, The dimensions and shape of the nanoparticles were also assessed via atomic force microscopy (AFM) (model Aist-NT-MDT, Russia), it was found that there was an irregular polygon shape of the silver particles with a diameter of 1.0–130 nm for the silver particles.

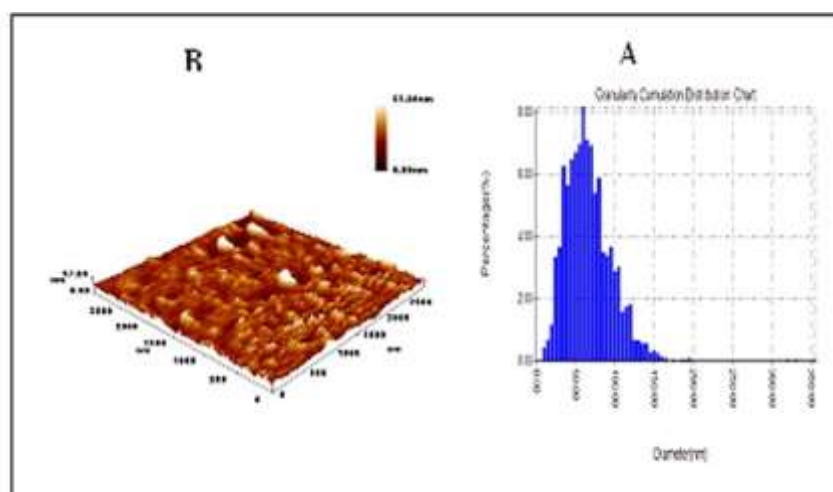


Figure 4.

3.6. Antibacterial Activity

The antibacterial activity of AgNPs was assessed against multiple pathogenic bacterial species, such as *Staphylococcus aureus*, *E. coli*, *Streptococcus pyogenes*, and *Klebsiella pneumoniae*, employing the agar well diffusion method. The inhibitory zones were strain-dependent, with measurements of 18.33, 12, 11.33, 9.66, and 9 ± 0.57 mm. The zone of inhibition was observed for *Staphylococcus aureus*, MIC and MBC value at $6.25 \mu\text{g/ml}$ followed by *Escherichia coli* with measurements of 15.66, 13.66, 12, 11, and 10 ± 0.87 mm, MIC and MBC value at $3.125 \mu\text{g/ml}$, then *Streptococcus pyogenes* with maximum measurement of 30.33, 16, 14, 11.33, and 10 ± 0.57 mm, MIC and MBC value at $3.125 \mu\text{g/ml}$, and *Klebsiella pneumoniae* with measurement of 20.33, 14, 12.33, 11.33, and 1 ± 0.57 mm at concentrations of 25, 12.5, and $6.25 \mu\text{g/ml}$, respectively MIC and MBC value at $6.25 \mu\text{g/ml}$ (Figure 5). The antimicrobial activity of nanoparticles of silver is determined by the dynamics between silver ions and biologically active compounds such as chamazulene, 1,8-cineole, camphor, and β -pinene of the essential oil of *A. millefolium* (Rathore et al., 2025). These compounds with silver nitrate destroy pathway against Gram-positive and Gram-negative bacteria. Owing to their smaller dimensions the nanoparticles have more bacterial cell membrane area for interaction, which triggers permeabilization and disruption of the membrane function and accelerates the cell death (Shami, 2025). Mechanism of action might involve of electrostatic attraction between the negative charge of the microbial cell membrane and the positive or smaller charge of AgNPs. The AMCSNPs were synthesized using 0.1% chitosan, 1% sodium tripolyphosphate, and 20% *A. millefolium*. The AMCSNPs had the greatest inhibitory effect on *Bacillus subtilis* (gram-positive) and *P. aeruginosa* (gram-negative) bacteria (30 ± 0.5) via the well-diffusion method, whereas, the greatest antialgal activity (68%) at the aggregate formation stage. Compare with gram-positive, the NPs demonstrated greater antibacterial efficacy against pathogenic and gram-positive bacteria and greater sensitivity because the peptidoglycan layer of Gram-positive bacteria acts as a physical filter that traps larger silver nanoparticles and limits their direct penetration into the cytoplasm. However, susceptibility is governed by a combination of physical trapping, electrostatic interaction, and ion release rather than penetration alone. The thick peptidoglycan layer of Gram-positive bacteria contains a high density of negatively charged teichoic acids (Gupta and Chauhan, 2021). This strongly attracts AgNPs and released silver ions (Ag^+) leading to massive accumulation on the cell surface. This localized concentration causes severe envelope stress and structural deformation without requiring immediate cytoplasmic penetration. In contrast, Gram-negative bacteria held a thin peptidoglycan layer and an outer membrane rich in lipopolysaccharides, which makes a negative surface charge that fascinates silver ions, promoting cell wall penetration and intracellular damage (Abd Qasim and Yaaqoob, 2023). This study suggest that *A. millefolium* nanoparticles may be a suitable substitute for chemicals as antibiotics, given the increasing trend of antibiotic resistance. This is due to the synergistic that take place between silver ions and bioactive molecules from essential oils on the surface of the nanoparticles that catalyze the antibacterial activity.

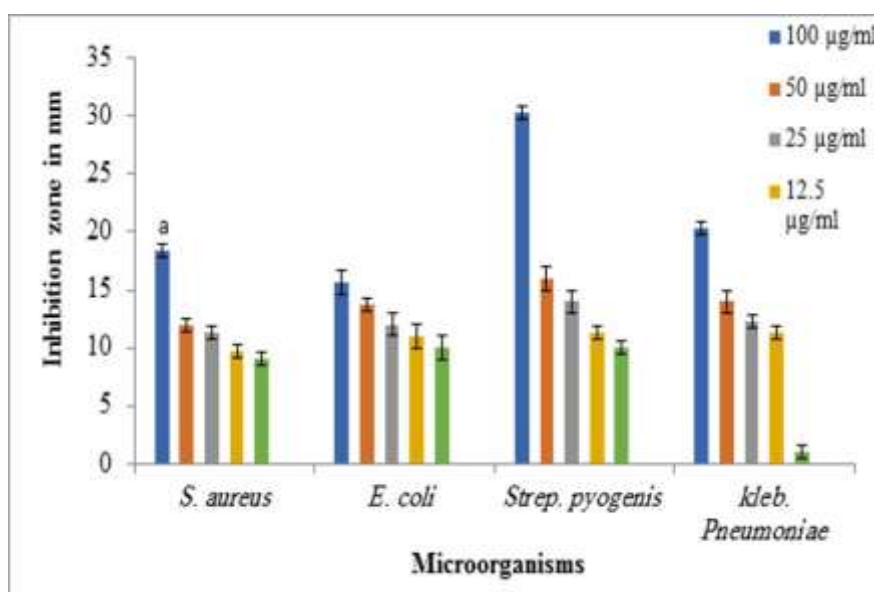


Figure 5.

3.7. Antioxidant Activity

This radical scavenging method was employed to assess the antioxidant ability of the resulting silver nitrate. The antioxidant effect of the NPS was good, as indicated by the DPPH free radical scavenging. The scavenging effect was dose dependent and 87.7, 77.4, 76.07, 74.46, and 71.01% were scavenged at concentrations of 100, 50, 25, 12.5, and 6.25 $\mu\text{g/ml}$, respectively compare with vitamin C (93.64%) (Figure 6). The activity was greater than that of the free essential oil of *A. millefolium* (60 $\mu\text{g/ml}$), demonstrating that the nanoparticles augment the antioxidant properties of the essential oil (Lasmi et al., 2025). The reducing capacity of the nanoparticles was significantly increased in a dose-dependent fashion, indicating that the particles can donate electrons to free radicals during a reaction. The reducing power of the nanoparticles was greater than that of the free essential oil and enhanced the antioxidant capacity of bioactive constituents from *A. millefolium* produced by the nanoparticles. One of the important mechanism of the essential oil of nanoparticles of silver nitrate of *A. millefolium* depend on chemical components such as flavonoids and terpenoids, with AgNO_3 donating electrons to stabilize harmful free radicals. Nanoencapsulation optimizes this process by controlling spatial orientation and managing electron transfer kinetics. The greater antioxidative effect of the nanoparticles is related to their greater surface area for feeding the free radicals. The essential oil of *A. millefolium* includes bioactive molecules (flavonoids and terpenoids) that have considerable antioxidant activity, and these compounds can be stabilized and improved by encapsulation in nanoparticles (Revathi et al., 2025). These findings agree with previous studies on the synthesis of plant-derived nanoparticles. which demonstrated that EO entrapment within nanocarriers resulted in improved antimicrobial and antioxidant potential (Shami, 2025). The application of green synthesis also leads to the environmental significance of this method, which prevents the use of toxic chemicals frequently applied in nanoparticle synthesis.

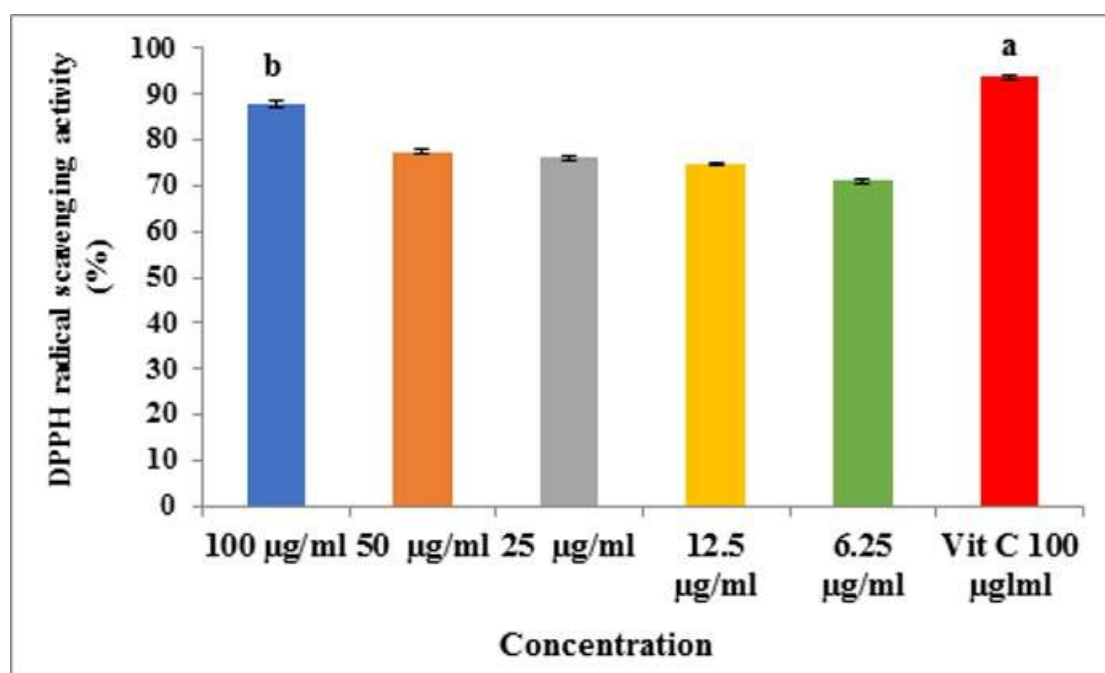


Figure 6.

4. CONCLUSION

The well-defined antibacterial and antioxidant effects of nanoencapsulated *A. millefolium* essential oils approach those of the unencapsulated essential oils. The environmentally sustainable production of silver nanoparticles utilizing *A. millefolium* essential oil represents a potential method for green manufacturing of silver nanoparticles. The structural characterization of the nanoparticles revealed that they were small, stable and spherical shape, possibly contributing to their bioactivity. The antibacterial effects of the nanoparticles were significant for the examined harmful bacteria, including both gram-positive and gram-negative strains. These results validate the possibility of applying these nanoparticles as green antimicrobial agents especially for antibiotic-resistant microorganisms. The considerable antioxidative activity exhibited by DPPH radical scavenging indicates that these nanoparticles effectively neutralize free radicals. The *A. millefolium* cache weed essential oil

nanoemulsion is promising as a safe alternative to synthetic antibacterial and antioxidant agents. Its dual activity proliferation within an environmentally friendly synthesis resulted in the choice of bioactive agents. These findings indicate limitations in the therapeutic efficacy associated with the choice of nanoparticle formulation, dose, and delivery route. Further in vivo investigations are needed to elucidate the safety and efficacy of these methods in practical applications.

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AUTHOR CONTRIBUTIONS

AbdulMuhsin M. Shami: Methodology, Laboratory Analysis. Data Curation, Formal Analysis. Supervision, Writing—Review & Editing.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICS STATEMENT

Ethical approval was not required for this study.

DATA AVAILABILITY STATEMENT

Data supporting the findings of this study are available from the corresponding author upon reasonable request.

REFERENCES

- [1] Liyana, E. P., Jasmine, J. S., 2025. Transformative potential of plant-based nanoparticles in cancer diagnosis and treatment: bridging traditional medicine and modern therapy. *Naunyn-Schmiedeberg's Archives of Pharmacology*. 16, 1-22.
- [2] Issa, R., Mahdi A., 2024. Chemical detection of some active compounds present in certain species of the genus *Achillea millefolium* from the Asteraceae family using hplc technology. *European Journal of Modern Medicine and Practice*. 4(7), 217-219.
- [3] Fantasma, F., Samukha, V., Saviano, G., Chini, M.G., Iorizzi, M., Caprari, C., 2024. Nutraceutical aspects of selected wild edible plants of the Italian central apennines. *Nutraceuticals*. 4(2), 190-231.
- [4] Konarska, A., Weryszko-Chmielewska, E., Sulborska-Różycka, A., Kieltyka-Dadasiewicz, A., Dmitruk, M., Gorzel M., 2023. Herb and flowers of *Achillea millefolium* subsp. *millefolium* L.: Structure and histochemistry of secretory tissues and phytochemistry of essential oils. *Molecules*. 28(23), 7791.
- [5] Haque, M., Jantan, I., Harikrishnan, H., Mohamed, M., 2019. Nanotechnology-based herbal medicines in relation to bioavailability and pharmacokinetic concepts. *Molecules* (2019) 24(3), 1-23.
- [6] Shami A.M., 2025. Antibacterial and antioxidant activities of nanoparticles of essential oil of *Hypericum perforatum*. *Advances in Natural Sciences: Nanoscience and Nanotechnology*. 16(4), 1-9.
- [7] Singh, P., Kim, Y., Zhang, D., Yang, D., Chin A., 2020. Biological synthesis of nanoparticles from plants and their potential as antimicrobial agents. *Journal of Integrative Medicine*. 26(9), 703.
- [8] Birusanti, A. B., Espenti, C. S., Surendra, T. V., Srinivas, B., Srinivasulu, M., Peddulaiah, K., Han S., 2025. Synthesis and characterization of multi-responsive iron oxide nanoparticles: Evaluation of antibacterial properties and photocatalytic activity. *Journal of Molecular Liquids*. 417, 9-17.
- [9] Sadeghi, B., Gholamhoseinpoor F., 2015. Study on the stability and green synthesis of silver nanoparticles using *Ziziphora tenuior* (Zt) extract at room temperature. *Journal of Nanostructure in Chemistry*. 5(3), 310-315.
- [10] Rathore, P., Swami, S., Meena, A., 2018. *Achillea millefolium*: A promising source of medicinal products with biological activities. *Journal of Pharmacognosy and Phytochemistry* 7(3), 140-1161.
- [11] Gupta, A., Chauhan N., 2021. Role of nanoparticles in enhancement of antioxidant activity: A review. *Biomedicine & Pharmacotherapy*. 138, 1-4.
- [12] Hamza, H., Nabila A., 2023. Nanoencapsulation of *Rosmarinus officinalis* essential oil into Chitosan crosslinked to Tripolyphosphate nanoparticles and Alginate/Chitosan nanoparticles: Formulation, characterization, in vitro release study, and in vivo evaluation. *Journal of Drug Delivery and Therapeutics*. (13(11), 45-55.

- [13] Shakeri, A., Sahebkar A. and Javadi B., 2016. *Melissa officinalis* L.—A review of its traditional uses, phytochemistry and pharmacology. *Journal of Ethnopharmacology*. 188, 204-228.
- [14] Abd Qasim, M., Yaaqoob, L., 2023. Evaluation of antibacterial activity of iron oxide nanoparticles synthesis by extracellular *Lactobacillus* against *Pseudomonas aeruginosa*. *Journal of Medicinal and Chemical Sciences*. 6, 1100-1111.
- [15] Elbossaty, W., 2017. Green tea as biological system for the synthesis of silver nanoparticles. *Journal of Biotechnology & Biomaterials*. 7, 3-5.
- [16] Chaudhuri, S.K., Chandela, S., Malodial, L., 2016. Plant Mediated Green Synthesis of Silver Nanoparticles Using *Tecomella undulata* Leaf Extract and Their Characterization. *Nano Biomedicine and Engineering*. 8(1), 1-8.
- [17] IS, M., Shami, A., Salih, H., 2024. Antibacterial and antioxidant activities of phenolic compounds from *Myrtus communis* callus. *Iraqi Journal of Agricultural Sciences*. 55(3), 1088-1097.
- [18] Shami, A., 2018. The Effect of Anthraquinones Fractions from *Morinda Citrifolia* Leaves against Pathogenic Bacteria with Antioxidant Activities. *Journal of Reports in Pharmaceutical Sciences*. (2018) 7(3) 231-238.
- [19] Shahzadi, S., Fatima, S., Shafiq, Z., Janjua, M.R., 2025. A review on green synthesis of silver nanoparticles (SNPs) using plant extracts: a multifaceted approach in photocatalysis, environmental remediation, and biomedicine. *RSC Advances*. 15(5), 3858-3903.
- [20] Tavallaei, O., Doostan, M., Khoshnevisan, K., Tahmasebi, A., Maleki, H., 2025. Wound healing promotion by poly (vinyl alcohol)/chitosan electrospun nanofibrous scaffold loaded with *Achillea wilhelmsii* extract. *Nanomedicine Journal*. 12, 110-121.
- [21] Amato, G., Adiletta, V., De Martino, L., Elshafie, H. S., Camele, I., Francolino, R., Feo, V. De, 2025. Eco-Sustainable management of weeds and phytopathogens using *Achillea millefolium* essential oil. *Industrial Crops and Products*. 235, 1-10.
- [22] Aćimović, M., Vujisić, L., Lončar, B., Ivanović S., 2025. Headspace Volatile Profiles of *Achillea* Species: *A. asplenifolia*, *A. crithmifolia*, *A. filipendulina*, and *A. virescens*. *Chemistry & Biodiversity*. 22, 2-9.
- [23] Bhat, M., Chakraborty, B., Kumar, R.S., Almansour, A.I., Arumugam, N., Kotresha, D., 2021. Biogenic synthesis, characterization and antimicrobial activity of *Ixora brachypoda* (DC) leaf extract mediated silver nanoparticles. *Journal of King Saud University – Science*. 33(2), 1-7.
- [24] Githala, C.K., Raj, S., Dhaka, A., Mali, S.C., Trivedi, R., Githala, CK, Raj, S., Dhaka, A, Mali, S.C., Trivedi, R. 2022. Phyto-fabrication of silver nanoparticles and their catalytic dye degradation and antifungal efficacy. *Frontiers in Chemistry*. 10, 1-17.
- [25] Lasmi, F., Hamitouche, H., Laribi-Habchi, H., Chafai, N., Lasmi, F., Hamitouche, H., Laribi-Habchi, H., Chafai, N., Silver nanoparticles (AgNPs), methods of synthesis, characterization, and their application: a review. *Plasmonics*, (2025) 20 1-34.
- [26] Revathi, S., Dey, N., Thomas, A., Issac, P., Shaik, M., Hussain, S., Guru A., 2025. Optimization of *Achillea millefolium*-Infused Chitosan Nanocarriers for Antibacterial and Dye Degradation Applications. *Chemistry & Biodiversity*. 22(4), e202402150.

Figure 1. UV-Visible absorption spectra of plant oil nanoparticles. which shows absorption peak at peak at 219 nm.

Figure 2. FTIR of nanoparticles of oil from plant revealed that strong absorption bands for chemical groups (OH, C=O- C-H, C=C, C-N).

Figure 3. GC-MS chromatogram of *A. millefolium* oil nanoparticles revealed main constituents are alpha-Pinene, 3-carene, eucalyptol, decane, thujone, camphor, endo-Borneol, and 1,3,8-p.beta-menthatriene. bourbonene, caryophyllene, beta-copaene, gamma-murolene, beta-guaiene, Humulene, beta-cubebene, gamma-murolene, triisopropylchlorosilane, beta-guaiene, camphene and 2,4-nonadienal.

Figure 4. Atomic force microscopy of plant oil (A): Three-dimensional image of *A. millefolium* oil (B): AFM image of size distribution of (*A. millefolium*) nanoparticles.

Figure 5. Inhibitory zones of *A. millefolium* oil nanoparticles against the indicated test microorganisms. Samples with a letters (a, b, c, d, and e) exhibit significant differences ($p < 0.05$) from one another and from the untreated group.

Figure 6. Antiradical activity against DPPH of *A. millefolium* oil nanoparticles. Various letters (a, b, c) represent a significant difference among samples ($p < 0.05$).