

ORIGINAL ARTICLE

In Vitro Evaluation of the Anti-Bacterial Activity of *Aloe Vera* and Mint Extract against Bacterial Isolates from Facial Acne

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ABSTRACT

Introduction: Acne is a common dermatological condition often associated with bacterial colonization, primarily *Staphylococcus aureus* and *Staphylococcus epidermidis*. The increasing resistance to conventional antibiotics has prompted the exploration of plant-based alternatives. This study investigates the antibacterial effects of Aloe vera extract and *Mentha spicata* essential oil on acne-associated bacterial isolates.

Method: A cross-sectional, laboratory-based study was conducted involving 51 facial acne samples. Bacterial isolates were identified using standard biochemical tests. *Mentha spicata* essential oil was obtained via hydro-distillation, while *Aloe vera* extract was prepared using hot air oven drying followed by dissolution in Dimethyl sulfoxide. The antibacterial activity was assessed using the agar well diffusion method.

Result: Out of 51 acne samples, 48 (94.11%) showed bacterial growth. *Staphylococcus epidermidis* was the most common isolate, followed by *Staphylococcus aureus* including Methicillin-resistant (MRSA) and Methicillin-sensitive (MSSA) strains. *Mentha spicata* essential oil exhibited an antibacterial effect against 81.13% of isolates (MIC and MBC: 25 µL/mL), while Aloe vera extract was active against 58.5% (MIC and MBC: 50 µL/mL).

Conclusion: Both *Aloe vera* and *Mentha spicata* demonstrated inhibitory effects against acne-associated bacteria. The stronger antibacterial activity of mint essential oil suggests its greater therapeutic potential in acne management.

Key words: Acne; *Aloe vera*; Essential oil; MRSA; MSSA; MIC; MBC

INTRODUCTION

Skin is the most exposed part of the human body, where different populations of microbes inhabit, reflecting their various niches¹. Many external factors such as temperature, humidity, and light exposure can alter the ecosystem of the skin, resulting in changes in microbial populations². Acne is a common non-infectious skin affliction frequently encountered, involving excess sebum production, modified lipid composition, duct blockage, bacterial colonization, and inflammation³. Acne develops when specialized follicles undergo pathological alterations, leading to the formation of non-inflammatory lesions (comedones) and inflammatory lesions (papules, pustules, and nodules), affecting mostly the face but also the back and chest⁴⁻⁵.

Heredity, hormones, nutrition, pollutants, climatic conditions, and bacterial species contribute to acne development, either alone or in combination⁶. Approximately 85% of adolescents and young adults are affected by this condition, which may occur at any age but is most prevalent at 14–17 years in females and 16–19 years in males¹. Presently, various acne treatments exist, including topical antibiotics, chemical peeling agents, oral antibiotics, retinoids, and hormones. However, bacterial resistance to different drugs has limited the choice of antibiotics for acne therapy^{5,7}. While pharmacotherapies for acne are effective, they are often associated with adverse effects⁸. Therefore, medicinal plants offer a novel alternative to antimicrobial medications, as they are naturally derived, less toxic, and associated with fewer adverse effects than synthetic pharmaceuticals⁹.

According to the World Health Organization, medicinal plants could be the best source of drugs. Almost 80% of the populations in developing countries rely on pre-existing knowledge of herbal medicines for the treatment of various diseases⁶. Plants and plant products are traditionally used for healing wounds, burns, fungal infections, viral infections, antibacterial and acaricidal activity against skin infections such as acne, herpes, and scabies, as well as for addressing inflammatory/immune disorders affecting the skin, tumor initiation, and promoting activity against skin cancer¹⁰⁻¹¹. *Aloe* is a genus containing about four hundred species of flowering succulent plants belonging to the Liliaceae family, which grow readily in hot and dry climates. *Aloe vera* is a typical xerophyte with thick, fleshy, strangely cuticularized spiny leaves¹². The name is derived from the Arabic word 'Alloeh,' which means 'bitter,' referring to the bitter taste of the liquid contained in the leaves¹³. *Aloe vera* poses a number of therapeutic uses, such as anti-inflammatory, immunostimulatory, antibacterial, antifungal, and cell growth stimulatory activities¹⁴. Collenchyma cells and thin-walled cells from the parenchyma of its leaves contain mucilaginous transparent gel, known as aloe vera gel. *Aloe vera* gel and its extracts have become popular and are generally recognized as safe substances for use in food, dietary supplements, and Ayurvedic medicines¹⁵.

Mentha (commonly known as mint or pudina) is a well-known genus in the family Lamiaceae, valued for its medicinal and aromatic properties. It is an important medicinal herb, both annual and perennial, with significant antimicrobial, antiviral, antioxidant, and antitumor activities, and exhibits some antiallergenic potential¹⁶⁻¹⁷. Medicinal aromatic plants,

defined as those that contain essential oils capable of volatilizing at room temperature, have been used since ancient times and are well known across civilizations for their nutritional, therapeutic, and cosmetic potential¹⁸. Essential oils (EOs) are volatile, natural, complex compounds characterized by a strong odor and are formed by aromatic plants as secondary metabolites⁵. EOs are stored in various parts of the plant (flowers, buds, leaves, fruits, seeds, bark, wood, roots), in varying quantities¹⁸. Although essential oils have a long history of pharmacological use, their role in the treatment of dermatological disorders remains unclear⁶. Many different disorders can be treated by medicinal plants found across the globe. Discovering the various natural chemicals available from plants and animals is necessary due to the increasing occurrence of bacterial resistance to antibiotics¹⁹. Medicinal plant oils provide a natural alternative to synthetic drugs, particularly against microbial agents. Essential oils derived from plants are safe and dependable, compared to costly synthetic drugs that have adverse effects²⁰. The herbal plants are in great focus as a remedy for skin diseases and infections as an alternative to antibiotics due to their innate antimicrobial action, natural origin, lower toxicity, safety, higher efficacy, multifunctionality, and minimal side effects²¹. *Aloe vera* possesses antibacterial, anti-inflammatory, and antioxidant properties that effectively combat acne-causing bacteria and soothe inflamed skin²². Meanwhile, mint leaves offer benefits for acne-prone skin as they contain salicylic acid and exhibit strong antibacterial properties, helping prevent acne by inhibiting bacterial growth and reducing inflammation²³. Hence, *Aloe vera* and mint are widely recognized for their medicinal properties, particularly in treating facial acne. Therefore, this study aims to determine the antibacterial potential of *Mentha spicata* essential oil and *Aloe vera* (L.) Burm.f. extract against bacteria isolated from acne.

METHODS

A cross-sectional laboratory-based study was conducted at the Department of Laboratory Medicine, Manmohan Memorial Institute of Health Sciences, Kathmandu, from September 2022 to April 2023. A total of 51 samples were collected aseptically from individuals presenting with facial acne lesions (Figure 1). Sample sites included the cheek, forehead, chin, jawline, nose, hairline, and eyebrow area.



Figure 1: Purulent acne

Isolation and identification of bacteria

The samples were inoculated on nutrient agar and blood

agar plates and incubated aerobically at 37°C for 24–48 hours. Bacterial isolates were identified based on colony morphology, Gram staining, and standard biochemical tests²⁴. The antibiotic sensitivity testing of isolates was performed by the Kirby–Bauer disk diffusion method on Mueller–Hinton agar (MHA) using standard methods recommended by CLSI guidelines (2012)²⁵. The antibiotics tested were amoxicillin (AMX), cloxacillin (COX), cefoxitin (CX), norfloxacin (NOX), gentamycin (GEN), amikacin (AK), novobiocin (NV), nitrofurantoin (NIT), erythromycin (E), clindamycin (CD), vancomycin (VA), chloramphenicol (C), linezolid (LZ) and meropenem (MRP).

Collection and identification of plants

Leaves of *Aloe vera* and mint were collected in September 2022 from Kirtipur Municipality, Kathmandu, Nepal. The approximate GPS coordinates for Kirtipur, in Degrees, Minutes, and Seconds (DMS) format, are 27° 38' 42.6" N latitude and 85° 15' 0.5" E longitude, situating the site within the Kathmandu Valley.

The collected plant materials were taxonomically identified at the National Herbarium and Plant Laboratories (NHPL), Godawari, Lalitpur, Nepal. A voucher specimen (Code No.: 128-2079/80) was deposited at the herbarium for future reference. The collection process complied with local guidelines, and all necessary permits were duly obtained.

Preparation of plant extracts.

Aloe vera (L.) Burm.f. extract preparation was carried out as described by Ramachandra and Rao²⁶. 800 gm of the parenchymatous tissue (gel) was scraped out using a sterile knife and placed in a sterile container. The gel was colorless and smooth in texture. The collected gel was placed on aluminum foil and dried in an oven at 80°C for 8 hours. After drying, the *Aloe vera* gel was removed and powdered using a grinder. A portion of 20 gm of the powdered gel was then dissolved in 200 mL of 10% DMSO and refluxed for 24 hours. The resulting content was filtered through Whatman filter paper No. 1 and stored as a stock solution in a container.

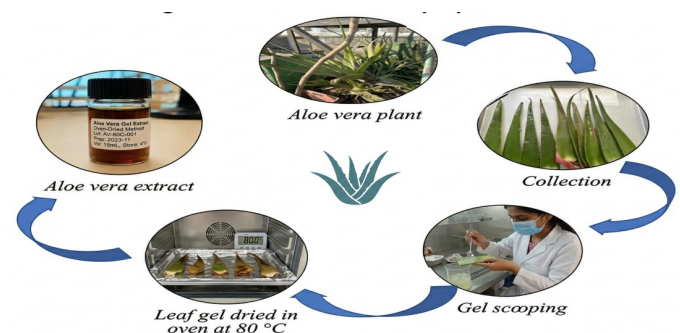


Figure 2: Aloe vera (L.) Burm.f. extract preparation

Mentha spicata essential oil preparation

Mentha spicata essential oil was prepared using the method described by Hazzit *et al.*²⁷, with minor adjustments. The fresh plant leaves were collected and thoroughly washed with water to remove dust. The leaves were then allowed to dry under shade for 40–50 days. After drying, the leaves were reduced to a powder using an electric grinder. For the extraction process, 50 gm of the powdered leaves were placed in a round-bottom flask with 600 mL of water and subjected to hydro-distillation using a Clevenger-type apparatus. The distilled essential oil was collected as a layer on top of the water in the graduated receiver, measured, and then dehydrated using anhydrous sodium sulfate for further use.



Figure 3: Essential oil preparation of mint by hydro distillation process

Determination of MIC and MBC

Mentha essential oil: The MIC of Mentha essential oil was determined using the broth dilution method. Two-fold serial dilutions (1.56–100 µL/mL) were prepared in Mueller-Hinton broth (MHB), and 0.1 mL of *Staphylococcus aureus* (ATCC 25923) was inoculated. After incubation at 37°C for 24 hr, MIC was recorded as the lowest concentration without turbidity²⁸. For MBC determination, aliquots from MIC and adjacent concentrations were plated on Mueller-Hinton agar (MHA) and incubated at 37°C for 24 hr. The lowest concentration showing no growth was recorded as MBC²⁹.

Aloe vera extract: MIC of *A. vera* extract was determined using the broth microdilution method in 96-well plates. Bacterial suspensions (adjusted to 0.5 McFarland standard) were diluted to 10⁶ CFU/mL in MHB. Two-fold dilutions of *A. vera* extract (1.56–100 µL/mL) were prepared, and 100 µL of bacterial suspension was added. After incubation at 37°C for 24 hr, MIC was determined by measuring absorbance at 490 nm³⁰. For MBC, aliquots from MIC and adjacent wells were plated on MHA and incubated at 37°C for 24 hr. The lowest concentration with no bacterial growth was recorded as MBC³¹.

Antibacterial activity testing

The antibacterial activity of the ethanolic extracts of *Aloe vera* and *Mentha* was evaluated using the cup plate method (Agar well diffusion assay)³². Isolates that produced a greater zone of inhibition (ZOI) than the reference strain *Staphylococcus aureus* ATCC 25923 were considered susceptible and thus active against the extract as described by Timilsena *et al.*³³.

The minimum inhibitory concentration (MIC) of the leaf extract was determined against the reference strain *Staphylococcus aureus* ATCC 25923 and used as a functional cutoff to assess the susceptibility of bacterial isolates. The corresponding ZOI at this MIC concentration served as a comparative benchmark. Isolates showing ZOI values greater than this threshold were classified as susceptible. This MIC-based interpretive approach allowed the reference strain to act as an internal standard, focusing the assessment on extract efficacy relative to a defined MIC threshold as described by Baral *et al.*³⁴.

A working solution of Mentha essential oil (25 µL/mL) was prepared by dissolving the oil in 50% ethanol. The solution was stored in tightly capped amber vials, sealed with adhesive foil, and refrigerated until use.

Preparation of bacterial suspension

Three to five well-isolated colonies of each test organism with similar morphology were picked using an inoculating wire and transferred into 5 mL of nutrient broth. The cultures were incubated at 37 °C for 2–4 hrs until the turbidity matched the 0.5 McFarland standard, equivalent to a bacterial suspension of 1 × 10⁸ – 2 × 10⁸ CFU/ML³⁵.

A sterile cotton swab was dipped into the bacterial suspension, rotated several times, and pressed firmly

against the inner wall of the tube to remove excess liquid. A Mueller-Hinton agar plate (4 mm thickness) was inoculated by swabbing the entire surface in three directions, rotating the plate 60° between streaks. The inoculum was allowed to absorb for 5–15 minutes with the lid in place.

Well preparation and sample application

Using a sterile borer (No. 6, 6 mm diameter), four wells were made in each MHA plate and labeled as follows:

- Control for essential oil: 50% ethanol
- Test for essential oil: 25 µL/mL essential oil
- Control for *Aloe vera* extract: 10% DMSO
- Test for *Aloe vera* extract: 50 µL/mL *Aloe vera* extract

Each well was filled using a micropipette. The plates were left undisturbed for 30 minutes to allow diffusion of the test substances into the agar medium. They were then incubated aerobically at 37 °C for 24 hrs in an upright position.

Measurement of ZOI

After incubation, the ZOI was measured in millimeters, including the diameter of the well. All tests were performed in triplicate to ensure reproducibility³⁰. Ciprofloxacin (5 µg/disc) was used as a positive control, while ethanol and DMSO served as negative controls.

Ethical Considerations

Ethical approval for this study was obtained from the Institutional Review Committee (IRC) of Manmohan Memorial Institute of Health Sciences (Ref no. 049/79), Kathmandu. Written informed consent was obtained from all participants prior to sample collection, in accordance with ethical guidelines for human research.

Data analysis

Data were analyzed using SPSS version 26.0. Descriptive statistics (frequency, percentage, mean ± SD) summarized specimen sites, bacterial isolates, and antibacterial activity. An independent t-test compared the mean zone of inhibition (ZOI) between the reference strain and acne-isolated bacteria for each extract. A p-value <0.05 was considered statistically significant. Antibiotic susceptibility was expressed as the percentage of sensitive and resistant isolates.

RESULTS

Among the 51 individuals with facial acne enrolled in the study for sample collection, the highest numbers of samples were collected from the cheek, while the lowest numbers of samples were collected from the hairline and eyebrow area (Table 1).

Table 1. Distribution of Specimen Site

Specimen site	Frequency (n)	Percent (%)
Hairline	1	2.0
Forehead	12	23.5
Eyebrow Area	1	2.0
Nose	8	15.7
Cheek	15	29.4
Jawline	5	9.8
Chin	9	17.6
Total	51	100.0

A total of 51 samples were analyzed, yielding 53 bacterial isolates. Mono-microbial growth of *Staphylococcus*

epidermidis was the predominant pattern observed in 28 samples (54.9%), followed by *Staphylococcus aureus* in 15 samples (29.4%). Bi-microbial growth was observed in 5 samples (9.8%), while 3 samples (5.9%) showed no growth. Among the total 53 isolates obtained, 33 were identified as *S. epidermidis* and 20 as *S. aureus*. Further testing of the 20 *S. aureus* isolates revealed that 4 were methicillin-resistant and 16 were methicillin-sensitive (Table 2).

Table 2. Pattern of bacterial growth (n = 51) and isolate characteristics(n = 53)

Pattern of Growth / Bacteria Isolated	Sample Frequency, n (%)	Total Isolates Yielded, n (%)	Strain Break-down
<i>S. epidermidis</i> only	28 (54.9)	33(62.3)	All <i>S. epidermidis</i>
<i>S. aureus</i> only	15 (29.4)	20(37.7)	4 MRSA, 16 MSSA
Bi-microbial growth	5 (9.8)	Accounted for above*	-
No growth	3 (5.9)	0	-
Total	51 (100.0)	53 (100)	-

*Note: The 5 bi-microbial samples yielded 5 additional *S. aureus* and 5 additional *S. epidermidis* isolates, bringing their respective isolate totals to 20 and 33.

Among the 53 bacterial isolates tested, Amikacin, Nitrofurantoin, and Meropenem were found to be absolutely sensitive (100%), followed by Gentamycin, Chloramphenicol, and Linezolid, which showed high sensitivity (98.1%). Other antibiotics such as Vancomycin (92.5%), Cefoxitin (90.6%), and Clindamycin (84.9%) also demonstrated strong effectiveness. On the other hand, Erythromycin exhibited the highest resistance (49.1%), followed by Cloxacillin (18.9%). The detailed antibiotic susceptibility pattern is shown in Table 3.

Table 3. Antibiotic susceptibility pattern of the isolates (n=53)

Antibiotic	Sensitive(%)	Resistant(%)
Amoxicillin (AMX)	51 (96.2)	2 (3.8)
Cloxacillin (COX)	43 (81.1)	10 (18.9)
Cefoxitin (CX)	48 (90.6)	5 (9.4)
Norfloxacin (NOX)	50 (94.3)	3 (5.7)
Gentamycin (GEN)	52 (98.1)	1 (1.9)
Amikacin (AK)	53 (100.0)	0 (0.0)
Novobiocin (NV)	50 (94.3)	3 (5.7)
Nitrofurantoin (NIT)	53 (100.0)	0 (0)
Erythromycin (E)	27 (50.9)	26 (49.1)
Clindamycin (CD)	45 (84.9)	8 (15.1)
Vancomycin (VA)	49 (92.5)	4 (7.5)
Chloramphenicol (C)	52 (98.1)	1 (1.9)
Linezolid (LZ)	52 (98.1)	1 (1.9)
Meropenem (MRP)	53 (100.0)	0 (0.0)

The percentage yield of mint essential oil was 3.14%. *Aloe vera* inhibited the growth of *Staphylococcus aureus* (ATCC 25923) at a MIC of 50 µL/mL, with no growth observed at the same concentration (MBC: 50 µL/mL). Similarly, mint essential oil exhibited a MIC of 25 µL/mL, with complete bacterial inhibition at MBC: 25 µL/mL (Table 4).

Table 4. MIC And MBC of plants extracts

Plant extracts	Bacterial strain	MIC	MBC
Aloe vera	ATCC 25923 (<i>S. aureus</i>)	50µL/mL	50µL/mL
Mint	ATCC 25923 (<i>S. aureus</i>)	25µL/mL	25µL/mL

Among the 53 isolates tested, mint essential oil exhibited the highest antimicrobial effect (81.13%), while Aloe vera extract showed the lowest effect (58.50%) (Table 5).

Table 5. Antibacterial effect of plant extracts on acne-isolated bacteria

S.N.	Plant Extract	Active (%)	Inactive(%)
1	<i>Aloe vera</i>	31 (58.50)	22 (41.50)
	<i>S. aureus</i>	9 (16.98)	11 (20.75)
	<i>S. epidermidis</i>	22 (41.51)	11 (20.75)
2	Mint	43 (81.13)	10 (18.87)
	<i>S. aureus</i>	16 (30.18)	4 (7.54)
	<i>S. epidermidis</i>	27 (50.95)	6 (11.33)

When a Student's t-test was used to compare the ZOI values between standard and acne-isolated bacteria for each extracts, the mean zone of inhibition (ZOI) for Aloe vera was 11.3 ± 0.5 mm against ATCC 25923 and 10.47 ± 2.43 mm against acne isolates, with no statistically significant difference observed (p = 0.287).

In contrast, *Mentha spicata* extract exhibited a mean ZOI of 12.4 ± 1.4 mm against ATCC 25923 and a significantly higher inhibition zone of 13.96 ± 2.06 mm against acne isolates (p = 0.034), indicating enhanced antibacterial activity against clinical isolates (Table 6). These findings suggest that *Mentha spicata* may have superior efficacy against acne-associated bacteria compared to the reference strain, while *Aloe vera* demonstrated relatively stable activity across both test groups.

Table 6. Mean ZOI against *Staphylococcus aureus* (ATCC 25923) and acne-isolated bacteria

Plant Extract	Mean ZOI – ATCC 25923 (mm)	Mean ZOI – Acne-isolated Bacteria (mm)	p-value
<i>Aloe vera</i>	11.3 ± 0.5	10.47 ± 2.43	0.287
<i>Mentha spicata</i> (Mint)	12.4 ± 1.4	13.96 ± 2.06	0.034*

*Significant (p < 0.05)

DISCUSSION

Acne is one of the most common skin disorders, primarily affecting adolescents. *Propionibacterium acnes* and *Staphylococcus epidermidis* are considered the major skin bacteria responsible for acne formation³⁶. Despite the wide range of anti-acne agents available, dermatologists continue to struggle with achieving successful treatment. Consequently, natural remedies are increasingly being explored as alternative treatments for acne.

This study was conducted among 51 individuals with acne, comprising 42 females and 9 males, which is consistent with findings by Kapoor and Saraf *et al.*, who reported that females are more prone to acne than males³⁷. The age range of individuals with acne in our study was 16–30 years. This descriptive cross-sectional study aimed to determine the antibacterial effect of *Aloe vera* and mint essential oil against bacteria isolated from facial acne. Among the

isolates, 20 were identified as *Staphylococcus aureus*, of which 4 were methicillin-resistant (*MRSA*) and 16 were methicillin-sensitive (*MSSA*). Additionally, 33 isolates were identified as *Staphylococcus epidermidis*.

The antibacterial effects of essential oils (EOs) are generally dependent on their chemical composition, which can be influenced by various factors such as the plant's developmental stage, the plant part used for extraction, geographical location, and soil and climate conditions³⁸. Consequently, the mint essential oil used in our study may have exhibited unique bioactive properties due to the fact that it was collected from Kirtipur, Nepal.

The use of *Mentha* essential oils as antibiotics or alternative treatments for infectious diseases has been suggested based on traditional medicinal practices, increasing antibiotic resistance, and the adverse effects of conventional antibiotics. Numerous plant extracts and essential oils have demonstrated antimicrobial properties and have been used topically as antiseptics. However, the potential of plant oils and extracts as sources of novel antibacterial substances requires further investigation³⁹.

The *in vitro* antibacterial activity of the dimethyl sulfoxide (DMSO) extract of *Aloe vera* leaf gel demonstrated varying degrees of inhibitory effects at different concentrations. The highest zone of inhibition (ZOI) against *S. aureus* was observed at 100, 200, and 400 µg/ml of DMSO extract, with ZOI measurements of 12 mm, 14 mm, and 16 mm, respectively. In our study, the mean ZOI was found to be 11.3 mm at a 50 µl/ml concentration, which is comparable to the 100 µg/ml concentration reported in the study by Haque *et al.*⁴⁰.

Kaithwas *et al.* reported mean inhibition zone diameters for *Aloe vera* gel of 9.56±0.89 mm against *S. aureus* and 6.00±0.00 mm against *S. epidermidis*, indicating sensitivity to *S. aureus* and resistance to *S. epidermidis*. The results in our study were slightly higher⁴¹. Similarly, Yousafzai *et al.* found that *Aloe vera* plant extracts were effective against acne and skin pimples, with different concentrations producing varying effects on skin lesions. At higher doses, *Aloe vera* facilitated complete healing of lesions, drying of pus-filled lesions, and disappearance of cysts. However, our study was limited to *in vitro* conditions, and further *in vivo* studies are required for better interpretation⁴².

Lawrence reported ZOI measurements of 15.66 mm and 14 mm against *S. aureus* (MTCC 2943) using ethanolic and methanolic extracts of *Aloe vera*, respectively. Our study reported lower ZOI values, likely due to the use of *S. aureus* (ATCC 25923) as the reference strain and the use of DMSO extract⁴³. In another study, methanolic extracts of *Aloe vera* pulp showed an 18 mm ZOI, while aqueous extracts of the leaf pulp showed no inhibition against *S. aureus*⁴⁴.

Essential oils from *Mentha* species have demonstrated

antibacterial activity in various studies. Essential oils from *Mentha* species from Spain and India, diluted in almond oil at 20% (v/v), showed ZOI values of 8.5±1.5 mm and 6.5±0.5 mm, respectively, against *S. epidermidis*. In contrast, our study used mint essential oil dissolved in 50% ethanol, resulting in a ZOI of 13.96±2.06 mm against *S. aureus*⁴⁵.

The oil yield of the Tunisian variety of spearmint was reported as 1.1%, which is lower than the spearmint used in our study. This difference may be due to geographical variation⁴⁶. Our study reported a *Mentha* essential oil yield of 3.14%, which was higher than the yield reported by *Spicata* (0.57%–1.4%)⁴⁷. Jeyakumar *et al.* reported that peppermint oil exhibited the highest activity against *S. aureus*, producing a maximum ZOI of 24.33 mm. In contrast, our study found a ZOI of 13.96 mm, which may be attributed to differences in the *Mentha* species used (*Mentha spicata* in our study)⁴⁸. Similarly, Zaidi and Dabiya *et al.* reported that *Mentha spicata* and *Mentha piperita* essential oils exhibited strong antibacterial activity against *S. aureus*, producing ZOI values of 21±0.09 mm and 19.2±0.07 mm, respectively. The differences in our findings may be due to variations in essential oil composition⁴⁹.

Ethyl acetate extracts of peppermint have demonstrated strong inhibitory effects against both *MRSA* and *MSSA*. Our study similarly concluded that *Mentha spicata* essential oil can inhibit the growth of both *MRSA* and *MSSA*⁵⁰. Othman and Kamel *et al.* reported that *M. spicata* essential oil demonstrated greater antimicrobial activity against Gram-negative bacteria than Gram-positive bacteria. However, in our study, only Gram-positive bacteria were isolated from acne samples, with an 81.13% effectiveness rate⁵¹.

This study had several limitations. First, the sample size was relatively small, and the study was conducted over a short duration, which may limit the statistical power and generalizability of the findings. Second, there may have been degradation of essential oils during extraction or storage, potentially affecting antibacterial activity. Lastly, plant samples were collected from a single geographical area, which may not represent the phytochemical diversity found in other regions of the country.

CONCLUSIONS

This study highlights the antibacterial potential of *Aloe vera* extract and *Mentha spicata* (mint) essential oil against *Staphylococcus aureus* and *Staphylococcus epidermidis*, bacteria commonly associated with facial acne. The findings suggest that the secondary metabolites present in these medicinal plants contribute to their antimicrobial properties, making them promising alternatives for acne treatment. Further research is necessary to isolate and identify the specific bioactive compounds responsible for these effects, particularly in *Aloe vera*, which could lead to the development of novel and more potent antimicrobial agents. Additionally, this study encourages further exploration of plant-based extracts as cost-effective, accessible, and sustainable solutions for dermatological applications.

REFERENCES

1. Smith L, Johnson T. Microbial populations of the skin and their role in human health. *J Dermatol Sci*. 2021;68(1):10-20.
2. Williams S, Lee B. The effect of environmental factors on skin microbial ecosystems. *Environ Microbiol*. 2020;32(5):220-227.
3. Patel A, Khan M. Acne vulgaris: Pathogenesis and treatment. *Dermatol Clin*. 2019;37(3):251-258.
4. Zhang X, Yu M, Chen Y, et al. Acne lesions and their impact on human skin. *J Clin Dermatol*. 2018;16(6):439-446.
5. Rogers M, Davis P. Acne vulgaris treatment and resistance. *Antibiotics Ther*. 2020;45(2):123-131.
6. Carter R, Brown S. The genetic and environmental factors in acne development. *Int J Dermatol*. 2019;58(9):1044-1050.
7. Lee K, Tan H. Bacterial resistance to common acne medications. *Clin Infect Dis*. 2021;62(1):35-42.
8. Smith J, Chen L. Adverse effects of acne pharmacotherapies. *J Dermatol Ther*. 2020;11(3):222-229.
9. Gupta N, Patel P. Medicinal plants in the treatment of acne: A natural alternative. *Phytomedicine*. 2020;25(4):365-370.
10. World Health Organization. WHO Traditional Medicine Strategy 2014–2023. Geneva: World Health Organization; 2013.
11. Gupta V, Rathi S. Herbal medicine in the management of acne. *J Cosmet Dermatol*. 2019;18(1):41-47.
12. Zhang Q, Li L, Xu C, et al. Herbal medicine for treating acne vulgaris: A review of clinical evidence and mechanisms. *Phytomedicine*. 2018;45:103-112.
13. Williams A, Harris R. Aloe vera: A comprehensive review of the biological properties. *J Ethnopharmacol*. 2018;225:218-229.
14. Eshun K, He Q. Aloe vera: A valuable ingredient for the food, pharmaceutical, and cosmetic industries—A review. *Crit Rev Food Sci Nutr*. 2004;44(2):91-96.
15. Arora A, Sharma N. Aloe vera and its therapeutic applications: A review. *Int J Pharm Sci Rev Res*. 2019;56(1):136-141.
16. Rivas A, Rocha E, Neves N. Aloe vera gel: Properties and applications. *Phytochem Rev*. 2017;16(3):451-463.
17. Mahmood N, Shaikh M. Medicinal properties of mint. *J Herb Med*. 2018;6(2):73-80.
18. Kumar A, Jaiswal S. Mint as a medicinal plant: A review. *Pharmacogn J*. 2019;11(2):358-366.
19. Battinelli L, Maggi F. Essential oils in herbal medicine: Volatile compounds and their therapeutic applications. *Med Aromat Plants*. 2020;5(1):10-18.
20. Usha M, Gopalakrishnan L. The rise of antibiotic resistance: A major challenge in modern medicine. *Indian J Med Microbiol*. 2017;35(2):160-170.
21. Palombo EA. Traditional medicinal plant extracts and natural products with activity against oral bacteria: potential application in the prevention and treatment of oral diseases. *Evid Based Complement Alternat Med*. 2011;2011:680354.
22. Surjushe A, Vasani R, Saple DG. Aloe vera: A short review. *Indian J Dermatol*. 2008;53(4):163-166.
23. Nayak S, Nalabothu P, Sandiford S, Bhogadi V, Adogwa A. Evaluation of wound healing activity of *Allamanda cathartica* L. and *Aloe vera* in rats. *BMC Complement Altern Med*. 2006;6:12.
24. Cheesbrough M. *District Laboratory Practice in Tropical Countries*. 2nd ed. Cambridge, UK: Cambridge University Press; 2006.
25. Clinical and Laboratory Standards Institute (CLSI). *Performance Standards for Antimicrobial Disk Susceptibility Tests*. Approved Standard, 7th ed. CLSI Document M02-A11. Wayne, PA: CLSI; 2012.
26. Ramachandra TV, Rao MR. Extraction and characterization of Aloe vera gel: Impact on properties. *Asian J Biol Sci*. 2008;2(3):502-510.
27. Hazzit M, Baaliouamer A, Faleiro ML, Miguel MG. Chemical composition, antioxidant, antimicrobial and cytotoxic activities of *Mentha spicata* L. essential oil and its phytochemicals menthol and carvone. *J Appl Microbiol*. 2006;100(6):1203-1210.
28. Wiegand I, Hilpert K, Hancock RE. Agar and broth dilution methods to determine the minimal inhibitory concentration (MIC) of antimicrobial substances. *Nat Protoc*. 2008;3(2):163-175.
29. Balouiri M, Sadiki M, Ibsouda SK. Methods for in vitro evaluating antimicrobial activity: A review. *J Pharm Anal*. 2016;6(2):71-79.
30. Andrews JM. Determination of minimum inhibitory concentrations. *J Antimicrob Chemother*. 2001;48(S1):5-16.
31. Clinical and Laboratory Standards Institute (CLSI). *Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; approved standard—10th ed*. CLSI Document M07-A10. Wayne, PA: CLSI; 2015.
32. Perez C, Pauli M, Bazerque P. An antibiotic assay by the agar-well diffusion method. *Acta Biol Med Exp*. 1990;15:113-115.
33. Timilsina RP, Baral SK, Dhakal A, Dhungana B, Acharya B. Antimicrobial Potential of Three Nepalese Medicinal Plants Against Multidrug Resistance *Escherichia coli* Isolates From Normal Individuals. *Sci World J*. 2024;2024:8031371. <https://doi.org/10.1155/tswj/8031371>
34. Baral SK, Bhasima S, Parajuli I, Manandhar KD, Poudel P. Anti-MDR *Escherichia coli* Activity, Phenolic and Flavonoid Content, and Antioxidant Potential of *Azadirachta indica* A. Juss Ethanolic Leaf Extract: An HR-LCMS-Based Profiling Study. *Sci World J*. 2025;2025:6593165. <https://doi.org/10.1155/tswj/6593165>
35. Clinical and Laboratory Standards Institute (CLSI). *Performance Standards for Antimicrobial Susceptibility Testing*. 30th ed. CLSI Supplement M100. Wayne, PA: CLSI; 2020.
36. Beylot C, Auffret N, Poli F, Claudel JP, Leccia MT, Del Giudice P, et al. *Propionibacterium acnes*: an update on

- its role in the pathogenesis of acne. *J Eur Acad Dermatol Venereol.* 2014;28(3):271-278.
37. Kapoor S, Saraf S. Assessment of anti-acne potential of herbal formulations. *Int J Pharm Sci Res.* 2011;2(7):1642-1648.
 38. Burt S. Essential oils: their antibacterial properties and potential applications in foods—a review. *Int J Food Microbiol.* 2004;94(3):223-253.
 39. Bassolé IH, Juliani HR. Essential oils in combination and their antimicrobial properties. *Molecules.* 2012;17(4):3989-4006.
 40. Haque MA, Jantan I, Harikrishnan H, Sher Mohamed F, Aluwi MF, Israf DA. Phytochemical and pharmacological profile of Aloe vera: Progress and prospect. *Evid Based Complement Alternat Med.* 2018;2018:5175872.
 41. Kaithwas G, Dubey J, Kushwaha P, Bhatia D, Majumdar DK. Anti-inflammatory and antimicrobial efficacy of Aloe vera gel in experimental models. *Asian Pac J Trop Med.* 2014;7(Suppl 1):S126-S128.
 42. Yousafzai ZA, Khan N, Ahmad S, Khan A. Antibacterial activity of Aloe vera against acne-inducing bacteria. *J Tradit Chin Med.* 2020;40(2):290-295.
 43. Lawrence R, Tripathi P, Jeyakumar E. Isolation, purification and evaluation of antibacterial agents from Aloe vera. *Braz J Microbiol.* 2009;40(4):906-915.
 44. Pandey R, Mishra A. Antibacterial activities of crude extract of Aloe barbadensis to clinically isolated bacterial pathogens. *Appl Microbiol Biotechnol.* 2010;85(3):1165-1175.
 45. Kalpna R, Pramila Y, Satish D. Comparative antibacterial activity of essential oils of Mentha species from Spain and India against clinical isolates. *Int J Pharm Pharm Sci.* 2017;9(2):45-50.
 46. Oumzil H, Ghouami S, Rhayour K, Ildrissi A, Fkih-Tétouani S, Faid M, et al. Antibacterial and antifungal activity of essential oils of Mentha suaveolens. *Phytother Res.* 2002;16(8):727-731.
 47. Jeyakumar E, Lawrence R, Tripathi P. Antimicrobial activity of peppermint (Mentha piperita) essential oil against clinical isolates of Staphylococcus aureus. *J Essent Oil Res.* 2011;23(3):25-30.
 48. Zaidi S, Dabiya S. Comparative antibacterial activity of Mentha spicata and Mentha piperita essential oils against pathogenic bacteria. *Pak J Pharm Sci.* 2019;32(5):2171-2176.
 49. Mirjana S, Živković J, Soković M, Glamočlija J, Čirić A, Grubišić D. Chemical composition and antimicrobial activity of Mentha spicata L. essential oil. *Arch Biol Sci.* 2014;66(4):1451-1457.
 50. Smith-Palmer A, Stewart J, Fyfe L. Antimicrobial properties of plant essential oils and essences against five important food-borne pathogens. *Lett Appl Microbiol.* 1998;26(2):118-122.
 51. Othman L, Kamel M, Al-Bakri AG, Bustanji Y. Antimicrobial activity of essential oils of Mentha spicata and Mentha piperita against multidrug-resistant bacteria. *J Med Microbiol.* 2019;68(9):1243-1252.

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CONFLICT OF INTEREST

The authors declare no competing interests

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