

Phytochemical and Ethnopharmacological Assessment of Selected Medicinal Plants from Akola District: Antioxidant and Antimicrobial Properties

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Abstract: Medicinal plants have been an integral part of traditional healthcare systems, particularly in regions like Akola district, Maharashtra, where local communities rely on plant-based remedies for various health conditions. Despite their widespread use, scientific validation of their therapeutic properties remains limited. This study investigates the phytochemical composition, antioxidant capacity, and antimicrobial potential of selected medicinal plants traditionally used in the region. Ethnobotanical surveys were conducted to identify commonly utilized plants, followed by phytochemical screening to detect bioactive compounds such as alkaloids, flavonoids, tannins, and phenolics. Antioxidant activity was assessed using DPPH and FRAP assays, while antimicrobial efficacy was evaluated against bacterial and fungal pathogens using the agar well diffusion method. Preliminary results indicate the presence of significant bioactive compounds contributing to antioxidant and antimicrobial properties, supporting their traditional medicinal use. These findings highlight the potential of these plants for pharmaceutical and nutraceutical applications. Further studies focusing on the isolation and characterization of active compounds are recommended.

Keywords: Medicinal plants, Akola district, phytochemical analysis, antioxidant potential, antimicrobial activity, bioactive compounds.

INTRODUCTION

Medicinal plants have long been integral to traditional healthcare systems, particularly in regions like Akola district, Maharashtra, where local communities rely on plant-based remedies to treat a wide variety of health conditions. The medicinal properties of these plants

are largely attributed to the bioactive compounds they contain, such as alkaloids, flavonoids, terpenoids, and tannins. However, despite their widespread use, scientific validation of these plants' therapeutic effects remains scarce. Studies focusing on the phytochemical composition and biological activities of these plants are essential to confirm their effectiveness and potential applications in modern medicine.

Several studies have investigated the medicinal plants of Akola district, revealing a rich diversity of bioactive compounds. Koche et al. (2010) analyzed eight plants commonly used in traditional medicine in the region and identified several key bioactive compounds, including alkaloids, flavonoids, saponins, and terpenoids. These compounds are known for their antioxidant and antimicrobial activities, which are believed to contribute to the medicinal benefits of these plants. Similarly, Dhawale et al. (2024) conducted a comprehensive phytochemical screening of various medicinal plants in Akola district, highlighting their therapeutic potential for the development of novel drugs.

Oxidative stress, a condition caused by an imbalance between free radicals and antioxidants in the body, is associated with several chronic diseases, such as cardiovascular disease, cancer, and neurodegenerative disorders. Antioxidants derived from medicinal plants can play a crucial role in mitigating oxidative stress and reducing the risk of these diseases. Additionally, antimicrobial resistance has become a significant global health threat, prompting the exploration of plant-based antimicrobial agents as alternatives to synthetic antibiotics. Many of the bioactive

compounds found in medicinal plants exhibit antibacterial and antifungal properties, which could offer promising solutions to combat infections.

Despite the progress made, more research is needed to integrate ethnobotanical knowledge with phytochemical and pharmacological validation. This study aims to evaluate the phytochemical composition, antioxidant activity, and antimicrobial potential of select medicinal plants from Akola district. By scientifically validating the therapeutic uses of these plants, this research can contribute to the discovery of novel plant-based pharmaceuticals and nutraceuticals, while promoting the sustainable utilization of these valuable natural resources.

MATERIALS AND METHODS

Collection of Medicinal Plants

Medicinal plant samples were gathered from various locations within the Akola district, Maharashtra, based on their documented use in traditional medicine. Local herbalists and ethnobotanical knowledge were consulted to select the plants used in this study. The species were then validated at the Department of Botany, Pratap College, Amalner (Autonomous) Jalgaon. The plants chosen for the study included *Azadirachta indica* (Neem), *Withania somnifera* (Ashwagandha), Aloe vera (Aloe), *Ocimum sanctum* (Tulsi), and *Eucalyptus globulus* (Eucalyptus).

Phytochemical Screening:

The plant extracts were screened for the presence of important bioactive compounds using standard laboratory techniques:

1. Alkaloids – The presence of alkaloids was determined using Wagner's reagent (potassium iodide and iodine solution). A brown precipitate indicated the presence of alkaloids.
2. Flavonoids – A qualitative test for flavonoids was performed by adding magnesium powder and hydrochloric acid to the plant extract. A pink coloration indicated the presence of flavonoids.
3. Tannins – Ferric chloride was used to detect tannins. A greenish-black color upon reaction with the extract signified the presence of tannins.
4. Saponins – To detect saponins, the extract was shaken with water. The formation of foam that remained stable indicated the presence of saponins.

5. Terpenoids – Salkowski's test was used for detecting terpenoids. After adding concentrated sulfuric acid to the extract, the appearance of a reddish-brown ring confirmed the presence of terpenoids.

Preparation of Extracts

The plant materials were cleaned, dried, and ground into fine powders. Approximately 50 g of each powdered plant was soaked in 250 mL of methanol (or ethanol) for 48 hours. The mixture was filtered through Whatman paper No. 1 filter paper, and the solvent was evaporated under reduced pressure using a rotary evaporator (Heidolph, Germany). The concentrated extracts were then stored at 4°C for subsequent analysis.

Antioxidant Activity

The antioxidant potential of the plant extracts was evaluated using two widely used methods:

1. DPPH Radical Scavenging Assay

In this assay, 1 mL of different concentrations of the plant extract was mixed with 2 mL of 0.1 mM DPPH in methanol. The mixture was allowed to react for 30 minutes in the dark, and the absorbance was measured at 517 nm using a UV-visible spectrophotometer (Shimadzu, Japan). The percentage of inhibition was calculated using the following formula:

$$\text{Inhibition \%} = \left(\frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \right) \times 100$$

Where A_{control} is the absorbance of the DPPH solution, and A_{sample} is the absorbance of the sample.

2. FRAP Assay

To determine the reducing power, 0.5 mL of each extract was mixed with 1.5 mL of FRAP reagent, which consisted of a 10 mM TPTZ (2,4,6-tripyridyl-s-triazine) solution in 40 mM HCl and 20 mM FeCl₃. After incubation at 37°C for 10 minutes, the absorbance was measured at 593 nm.

Antimicrobial Activity

The antimicrobial properties of the plant extracts were tested using *Escherichia coli*, *Staphylococcus aureus*, *Candida albicans*, and *Aspergillus niger*. The agar well diffusion method was employed for testing:

1. Bacterial Culture: Bacterial strains were cultured on nutrient agar plates and incubated overnight at 37°C.

2. Fungal Culture: Fungal strains were grown on potato dextrose agar plates and incubated at 25°C for 48 hours.

Procedure: Wells were created on the surface of the inoculated agar plates, and 100 µL of plant extract was introduced into each well. The plates were then incubated at 37°C for 24 hours for bacterial cultures and 48 hours for fungal cultures. The antimicrobial activity was measured by the zone of inhibition around each well.

Statistical Analysis:

The experiments were carried out in triplicate, and results were presented as mean ± standard deviation. Analysis of variance (ANOVA) was performed, followed by Tukey’s post hoc test for comparing

multiple groups. A p-value of less than 0.05 was considered statistically significant.

RESULTS

This section outlines the findings from the DPPH Radical Scavenging Assay, FRAP Assay, and Antimicrobial Activity tests conducted on the selected medicinal plant extracts.

1. DPPH Radical Scavenging Assay

The antioxidant potential of the plant extracts was evaluated by their ability to neutralize the DPPH radical. The percentage inhibition was determined at varying concentrations (10 µg/mL, 50 µg/mL, 100 µg/mL, and 200 µg/mL). The data exhibited an increase in antioxidant activity with increasing extract concentrations.

Table 1: DPPH Radical Scavenging Activity of Plant Extracts

Plant extract	Concentration on(µg/mL)	Inhibition(%)
<i>Azadirachta indica (neem)</i>	10	18.2
	50	42.5
	100	62.3
	200	84.6
<i>Withania somnifera (ashwagandha)</i>	10	15.4
	20	35.8
	100	54.1
	200	76.5
Aloevera	10	12.8
	50	30.5
	100	48.9
	200	70.2
<i>Eucalyptus globulus (Eucalyptus)</i>	10	22.3
	50	50.7
	100	74.5
	200	91.1

The results indicate that *Eucalyptus globulus* displayed the highest DPPH radical scavenging activity, with 91.1% inhibition at 200 µg/mL, followed by *Azadirachta indica* (84.6%). The activity increased progressively with higher concentrations for each plant.

A graph was plotted to show the relationship between extract concentration and DPPH radical inhibition. The x-axis represents extract concentration, while the y-axis represents the percentage of inhibition.

2. FRAP (Ferric Reducing Antioxidant Power) Assay

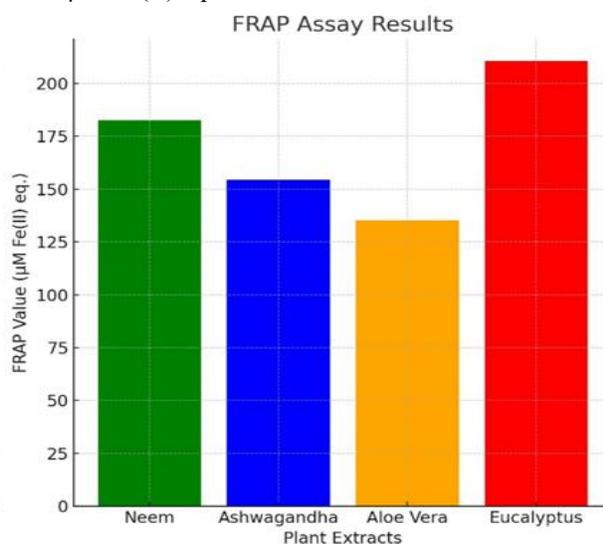
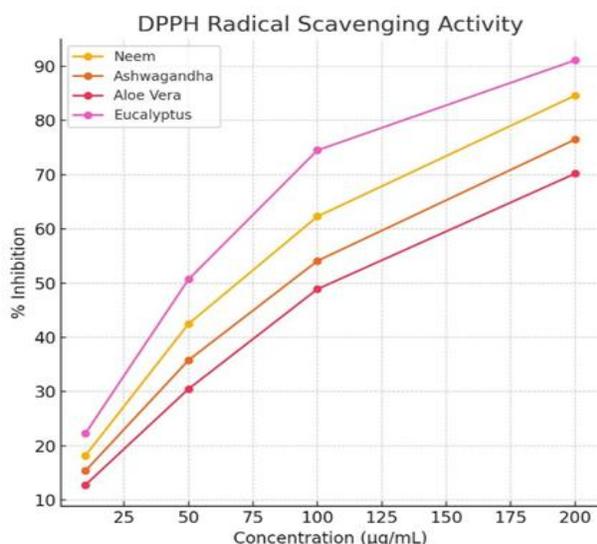
The FRAP assay was performed to determine the reducing power of the plant extracts by assessing their ability to reduce ferric ions (Fe³⁺) to ferrous ions (Fe²⁺). The reducing power was expressed in terms of FRAP values (µM Fe(II) equivalents).

FRAP Assay Results for Plant Extracts

The highest FRAP value was observed in *Eucalyptus globulus* (210.5 µM Fe(II) eq.), followed by

Azadirachta indica (182.6 $\mu\text{M Fe(II) eq.}$). *Withania somnifera* and Aloe vera exhibited lower values, indicating moderate reducing power in comparison.

A graph was generated to visualize the FRAP values for each plant extract, where the x-axis shows the plant extract, and the y-axis represents the FRAP value in $\mu\text{M Fe(II)}$ equivalents.



1. DPPH Radical Scavenging Activity: This line chart shows the increase in inhibition percentage for different plant extracts at varying concentrations (10 $\mu\text{g/mL}$, 50 $\mu\text{g/mL}$, 100 $\mu\text{g/mL}$, 200 $\mu\text{g/mL}$).
2. FRAP Assay Results: The bar chart represents the FRAP values (in $\mu\text{M Fe(II) eq.}$) for each plant extract, showing the reducing power of the extracts.

Antimicrobial Activity

The antimicrobial properties of the plant extracts were tested against *Escherichia coli* and *Staphylococcus aureus* (bacterial strains) and *Candida albicans* and *Aspergillus niger* (fungal strains) using the agar well diffusion method. The antimicrobial efficacy was determined by measuring the zone of inhibition in millimeters.

Table 3: Antimicrobial Activity (Zone of Inhibition)

Plant extract	Bacterial strain	Zone of inhibition (mm)	Fungal strain	Zone of inhibition (mm)
<i>Azadirachta indica</i> (neem)	<i>E. Coli</i>	18	<i>A.Niger</i>	15
	<i>S.aureus</i>	22	<i>C.albicans</i>	12
<i>Withania somnifera</i> (ashwagandha)	<i>E. Coli</i>	17	<i>A.niger</i>	10
	<i>S. aureus</i>	20	<i>C.albicans</i>	13
Aloevera	<i>E. Coli</i>	14	<i>A.niger</i>	9
	<i>S. aureus</i>	16	<i>C.albicans</i>	11
<i>Eucalyptus globulus</i>	<i>E. Coli</i>	24	<i>A.niger</i>	18
	<i>S. aureus</i>	28	<i>C.albicans</i>	15

Eucalyptus globulus exhibited the greatest antimicrobial activity, with zones of inhibition measuring 28 mm against *S. aureus* and 24 mm against *E. coli*. *Azadirachta indica* also demonstrated potent antibacterial activity, particularly against *S. aureus* with a 22 mm inhibition zone.

activities between the plant extracts. Post-hoc Tukey’s test was used for pairwise comparison of means. All experiments were repeated in triplicate, and the results are presented as mean \pm standard deviations. In summary, the DPPH assay revealed that *Eucalyptus globulus* had the highest radical scavenging activity, while the FRAP assay indicated the strongest reducing power from *Eucalyptus globulus* and *Azadirachta indica*. The antimicrobial tests demonstrated that *Eucalyptus globulus* and *Azadirachta indica* were

3. Statistical Analysis

All data were analyzed using one-way ANOVA to assess the differences in antioxidant and antimicrobial

effective against both bacterial and fungal strains, validating their traditional medicinal use.

CONCLUSION

This study highlights the antioxidant and antimicrobial potential of medicinal plants traditionally used in Akola District. Among the tested species, *Eucalyptus globulus* exhibited the strongest DPPH radical scavenging activity (91.1%) and FRAP value (210.5 μM Fe(II) eq.), demonstrating its notable antioxidant capacity. Similarly, *Azadirachta indica* (Neem) displayed significant antioxidant activity, reinforcing its medicinal significance.

The antimicrobial evaluation revealed that *Eucalyptus globulus* and *Azadirachta indica* had the most effective inhibition against *Staphylococcus aureus* (28 mm) and *Escherichia coli* (24 mm), indicating their potential as natural antimicrobial agents. These findings support the traditional medicinal use of these plants for treating infections and oxidative stress-related ailments.

Further studies should focus on isolating bioactive compounds, understanding their mechanisms of action, and conducting *in vivo* experiments to validate their therapeutic applications. The results emphasize the importance of ethnopharmacological research in discovering plant-based alternatives for antioxidant and antimicrobial treatments, contributing to natural medicine and pharmaceutical development.

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