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Antioxidant and Antimicrobial properties of *Rosmarinus officinalis* L. leaves

Chitra V[1], Hemalatha M[2], Yuvaraj M[3], Jayakumar S[4*]

[1]Assistant Professor of Chemistry, Dept of Biochemistry, SRM Arts and Science College, Kattankulathur, Tamilnadu, India.

[234]Assistant Professors, Dept of Biochemistry, SRM Arts and Science College, Kattankulathur, Tamilnadu, India.

*Corresponding author: Dr. S. Jayakumar

Abstract

The present study aimed to evaluate the antioxidant and antimicrobial properties of *Rosmarinus officinalis* L. leaf extracts. *R. officinalis* is a medicinal plant traditionally used for treating infections and oxidative stress–related disorders, largely due to its rich phytochemical composition. Methanolic and aqueous leaf extracts were prepared and assessed for antioxidant activity using enzymatic (catalase and superoxide dismutase) and non-enzymatic (ascorbic acid and vitamin E) assays. Antimicrobial activity was evaluated against *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, and *Pseudomonas aeruginosa* using the agar well diffusion method at varying extract concentrations. The methanolic extract of *Rosmarinus officinalis* L. leaves exhibited significantly higher antioxidant activity than the aqueous extract, with maximum catalase activity of 83.7 mg/g% and superoxide dismutase activity of 0.68 mg/g%**.** Non-enzymatic antioxidant levels were also highest in the methanolic extract, showing 76.24 mg/g% of ascorbic acid and 18.0 mg/g% of vitamin E. In antimicrobial assays, the methanolic extract demonstrated pronounced inhibitory activity at 100 µL, with maximum zones of inhibition of 6 mm against *Escherichia coli* and *Klebsiella pneumoniae*, and 5 mm against *Pseudomonas aeruginosa* and *Staphylococcus aureus* The results confirm that *R. officinalis* leaves possess strong antioxidant and antimicrobial potential, supporting their traditional use and suggesting their applicability in natural therapeutic formulations.

Keywords: *Rosmarinus officinalis,* Antioxidant activity, Antimicrobial activity, Medicinal plant

Introduction

Oxidative stress and microbial infections are major contributors to the development of various chronic and infectious diseases. Reactive oxygen species (ROS), when produced in excess, can damage cellular components such as lipids, proteins, and nucleic acids, leading to aging and pathological conditions including cancer, cardiovascular diseases, and neurodegenerative disorders. Antioxidants play a crucial role in neutralizing free radicals and protecting biological systems from oxidative damage. Similarly, the increasing emergence of antibiotic-resistant microorganisms has created an urgent need for alternative antimicrobial agents derived from natural sources.

Medicinal plants have long been used as sources of bioactive compounds with antioxidant and antimicrobial properties. According to the World Health Organization, a large proportion of the global population relies on plant-based medicines for primary healthcare. The growing interest in natural antioxidants and antimicrobials is largely due to their effectiveness, lower toxicity, and reduced side effects compared to synthetic compounds (WHO, 2000). Plant secondary metabolites such as phenols, flavonoids, alkaloids, tannins, and terpenoids are known to contribute significantly to these biological activities (Harborne, 1998). The family Lamiaceae includes several aromatic plants that are rich in phenolic compounds and essential oils, many of which exhibit strong antioxidant and antimicrobial properties. *Rosmarinus officinalis* L., commonly known as rosemary, is an evergreen perennial shrub native to the Mediterranean region and widely cultivated across the world. Rosemary leaves are extensively used in traditional medicine, food preservation, and cosmetic formulations. The plant has been traditionally employed to treat inflammatory disorders, digestive problems, headaches, infections, and nervous system ailments.

Previous studies have reported that rosemary extracts exhibit significant antioxidant activity due to the presence of phenolic diterpenes, flavonoids, and phenolic acids, which act as effective free radical scavengers (Rice-Evans et al., 1997). Rosemary has also been shown to possess antimicrobial activity against a wide range of pathogenic bacteria and fungi, supporting its traditional use as a natural preservative and therapeutic agent (Bakkali et al., 2008). The antimicrobial effects are often attributed to the ability of phytochemicals to disrupt microbial cell membranes and interfere with metabolic pathways. Despite numerous reports on the medicinal properties of rosemary, variations in extraction methods and solvent polarity significantly influence the biological activity of plant extracts. Therefore, systematic evaluation of antioxidant and antimicrobial properties using standardized assays is essential to validate its therapeutic potential.

In this context, the present study aims to evaluate the antioxidant and antimicrobial activities of *Rosmarinus officinalis* L. leaf extracts using enzymatic and non-enzymatic antioxidant assays and in vitro antimicrobial screening. The study seeks to provide scientific evidence supporting the traditional medicinal use of rosemary and to explore its potential application as a natural antioxidant and antimicrobial agent.

Materials and Methods

Plant Material Collection and Preparation

Fresh and healthy leaves of *Rosmarinus officinalis* L. were collected from cultivated plants and authenticated based on morphological characteristics. The collected leaves were washed thoroughly with distilled water to remove surface impurities and shade-dried at room temperature. The dried leaves were ground into a fine powder using a sterile grinder and stored in airtight containers for further analysis.

Preparation of Leaf Extracts

Leaf powder (100 g) was extracted separately using methanol and distilled water by maceration. Each solvent was added to the powdered material in a conical flask and kept at room temperature for 24 h with intermittent shaking. The extracts were filtered using Whatman No. 1 filter paper and concentrated using an air condenser. The crude extracts were stored at 4 °C until further use.

Antioxidant Activity Assays

The antioxidant activity of *Rosmarinus officinalis* L. leaf extracts was evaluated using enzymatic and non-enzymatic antioxidant assays. Catalase activity was determined following Sinha's method by measuring the decomposition of hydrogen peroxide spectrophotometrically. Superoxide dismutase (SOD) activity was assessed using the method described by Misra and Fridovich, based on the inhibition of epinephrine autoxidation. Non-enzymatic antioxidants, including ascorbic acid (vitamin C) and tocopherol (vitamin E), were estimated using standard colorimetric methods.

All measurements were performed in triplicate, and antioxidant activity was expressed as mg/g of extract (Sinha, 1972; Misra and Fridovich, 1972).

Antimicrobial Activity

The antimicrobial activity of *Rosmarinus officinalis* L. leaf extracts was assessed using the agar well diffusion method against selected Gram-positive and Gram-negative bacterial strains, namely *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, and *Pseudomonas aeruginosa*. Fresh bacterial cultures were prepared and standardized before inoculation. Sterile nutrient agar plates were uniformly swabbed with the test organisms, and wells were aseptically punched into the agar. Methanolic and aqueous plant extracts were tested at concentrations of 10, 20, 50, and 100 µL. Plates were incubated at 37 °C for 24 h, and antimicrobial activity was determined by measuring the zones of inhibition in millimeters (Perez et al., 1990; Balouiri et al., 2016).

Results

Enzymatic Antioxidant Activity

The Fig 1 represents the enzymatic antioxidant activity of R. officinalis **leaf extracts.** Catalase activity analysis revealed that the methanolic extract possessed significantly higher total catalase content (**83.7 mg/g%**) compared to the aqueous extract (**66.7 mg/g%**). Catalase plays a vital role in protecting cells from oxidative damage by decomposing hydrogen peroxide into water and oxygen, thereby preventing the accumulation of reactive oxygen species. Similarly, superoxide dismutase (SOD) activity was higher in the methanolic extract (**0.68 mg/g%**) than in the aqueous extract (**0.46 mg/g%**). The higher enzymatic antioxidant activity observed in the methanolic extract suggests its superior free-radical scavenging ability.

Non-enzymatic Antioxidant Activity

The non-enzymatic antioxidant parameters further supported the superior antioxidant capacity of the methanolic extract and the result was given in Fig 2. Total ascorbic acid content was found to be **76.24 mg/g%** in the methanolic extract, whereas the aqueous extract showed **62.17 mg/g%**. Vitamin E content followed a similar trend, with the methanolic extract exhibiting higher levels (**18.0 mg/g%)** compared to the aqueous extract (**16.02 mg/g%**) .The enhanced non-enzymatic antioxidant activity in the methanolic extract may be attributed to better solubility and extraction of antioxidant compounds in methanol.

Fig 1: Total catalase and SOD contents of *R. officinalis* leaf extracts

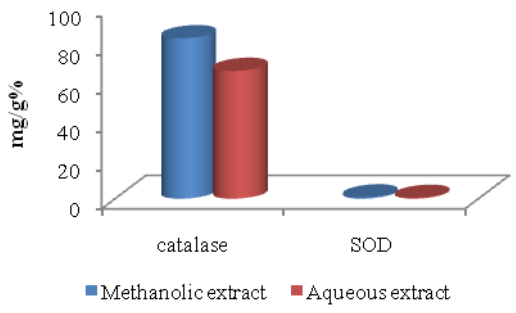
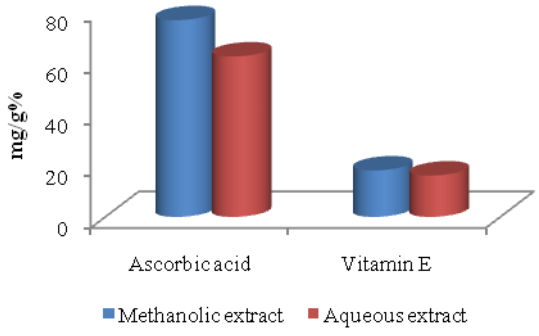


Fig 2: Total ascorbic acid and Vitamin E contents of *R. officinalis* leaf extracts



Antimicrobial Activity of *Rosmarinus officinalis* L. Leaves

The antimicrobial activity of the methanolic extract of *R. officinalis* leaves was evaluated against selected Gram-negative and Gram-positive bacterial strains using different concentrations (10, 20, 50, and 100 µL). The results demonstrated a concentration-dependent increase in antibacterial activity. Against *Escherichia coli*, inhibition zones increased from **2 mm at 10 µL** to **6 mm at 50 and 100 µL** (Fig. 3). Similarly, *Klebsiella pneumoniae* showed gradual increases in inhibition, reaching a maximum of **6 mm at 100 µL** (Fig. 4). In the case of *Pseudomonas aeruginosa*, the methanolic extract exhibited inhibition zones ranging from **2 mm (10 µL) to 5 mm (100 µL)** (Fig. 5). *Staphylococcus aureus* also showed notable susceptibility, with inhibition zones increasing from **1 mm at 10 µL to 5 mm at 100 µL** (Fig 6).

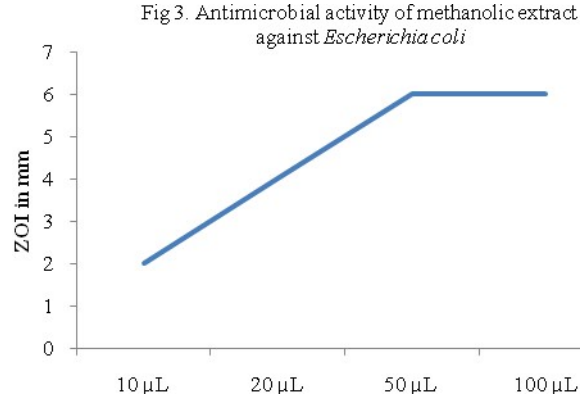


Fig 3. Antimicrobial activity of methanolic extract against *Escherichia coli*

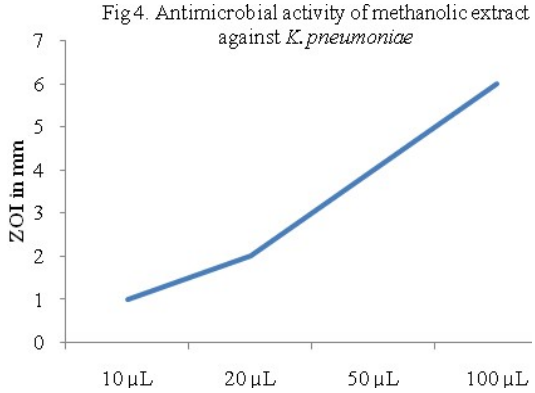


Fig 4. Antimicrobial activity of methanolic extract against *K. pneumoniae*

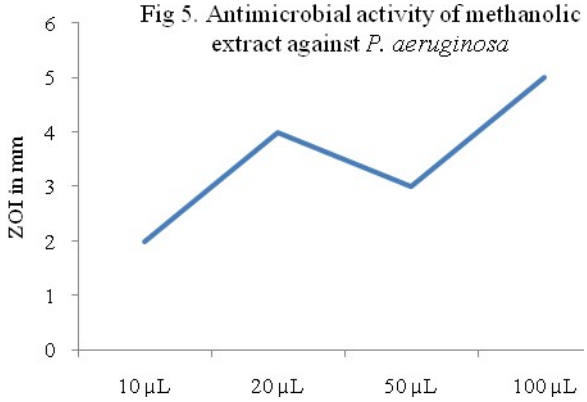


Fig 5. Antimicrobial activity of methanolic extract against *P. aeruginosa*

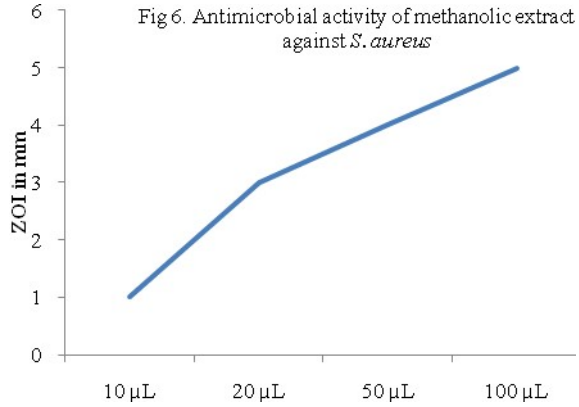


Fig 6. Antimicrobial activity of methanolic extract against *S. aureus*

Discussion

The present study demonstrates that *Rosmarinus officinalis* L. leaves possess considerable antioxidant and antimicrobial potential, with the methanolic extract consistently exhibiting superior activity compared to the aqueous extract. This variation highlights the significant influence of solvent polarity on the extraction efficiency of bioactive phytochemicals. Methanol is known to effectively extract phenolic compounds, flavonoids, and terpenoids, which are major contributors to antioxidant and antimicrobial activities in medicinal plants (Harborne, 1998; Sasidharan et al., 2011).

The enzymatic antioxidant assays revealed higher catalase and superoxide dismutase (SOD) activities in the methanolic extract than in the aqueous extract. Catalase plays a crucial role in cellular defense by decomposing hydrogen peroxide into water and oxygen, thereby preventing oxidative damage (Chitra *et al*., 2017). Similarly, SOD enzymes provide the first line of defense against reactive oxygen species by catalyzing the dismutation of superoxide radicals (Selvi *et al*., 2018). The elevated levels of these enzymes observed in the methanolic extract indicate a strong free-radical scavenging capacity of *R. officinalis* leaves.

Non-enzymatic antioxidants such as ascorbic acid and vitamin E were also found to be higher in the methanolic extract. Ascorbic acid is known to directly scavenge reactive oxygen species and regenerate other antioxidants, while vitamin E protects membrane lipids from peroxidation (Rice-Evans et al., 1997). The presence of these antioxidants further strengthens the antioxidant potential of rosemary leaves and supports previous reports highlighting rosemary as a potent natural antioxidant source (Aruoma et al., 1992).

The antimicrobial activity results demonstrated a concentration-dependent inhibitory effect of the methanolic extract against both Gram-negative and Gram-positive bacteria. *Escherichia coli* and *Klebsiella pneumoniae* were more susceptible, showing maximum zones of inhibition, while *Pseudomonas aeruginosa* and *Staphylococcus aureus* exhibited moderate sensitivity. These findings are consistent with earlier studies reporting the antibacterial properties of rosemary extracts and essential oils (Bakkali et al., 2008). The antimicrobial activity may be attributed to the presence of terpenoids and phenolic diterpenes such as terpineol, phytol, and ferruginol, which disrupt microbial cell membranes and interfere with essential metabolic processes (Yogananth et al., 2012).

Conclusion

The present investigation confirms that *Rosmarinus officinalis* L. leaves possess significant antioxidant and antibacterial potential. Both methanolic and aqueous extracts exhibited measurable antioxidant activity; however, the methanolic extract consistently demonstrated superior enzymatic and non-enzymatic antioxidant levels, including higher catalase, superoxide dismutase, ascorbic acid, and vitamin E contents. The methanolic extract also showed concentration-dependent antibacterial activity against both Gram-negative and Gram-positive bacteria, with maximum inhibition observed against *Escherichia coli* and *Klebsiella pneumoniae*. These findings highlight the influence of solvent polarity on the extraction of bioactive compounds and support the traditional medicinal use of rosemary. Overall, *R. officinalis* leaves represent a promising natural source of antioxidant and antimicrobial agents with prospective applications in pharmaceutical and nutraceutical formulations.

References

Aruoma, O. I., Halliwell, B., Aeschbach, R., and Loliger, J. (1992). Antioxidant and pro-oxidant properties of active rosemary constituents: Carnosol and carnosic acid. *Food and Chemical Toxicology*, 30(4), 351–356. https://doi.org/10.1016/0278-6915(92)90004-4

Balouiri, M., Sadiki, M., and Ibnsouda, S. K. (2016). Methods for in vitro evaluating antimicrobial activity: A review. *Journal of Pharmaceutical Analysis*, 6(2), 71–79. https://doi.org/10.1016/j.jpha.2015.11.005.

Bakkali, F., Averbeck, S., Averbeck, D., and Idaomar, M. (2008). Biological effects of essential oils—A review. *Food and Chemical Toxicology*, 46(2), 446–475. https://doi.org/10.1016/j.fct.2007.09.106

Chitra J., Syed Ali, M., Anuradha,V. and Yogananth, N. (2017). Quantification of free radical scavenging potential and antioxidant activity of methanolic extract from *Rhizophora mucronata* Bark. *International Journal of Advanced Biotechnology and Research*. 8(4): 463-478.

Harborne, J. B. (1998). *Phytochemical methods: A guide to modern techniques of plant analysis* (3rd ed.). Springer.

Selvi, R., Yogananth N., Syed Ali M. and Anuradha, V. (2018). *In vitro* antioxidant and free radical scavenging activity of crude extract of *Solanum nigrum* Linn. *International journal of scientific research and review*. 7(3): 153-163.

Misra, H. P., and Fridovich, I. (1972). The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. *Journal of Biological Chemistry*, 247(10), 3170–3175.

Perez, C., Pauli, M., and Bazerque, P. (1990). An antibiotic assay by the agar well diffusion

method. *Acta Biologiae et Medicinae Experimentalis*, 15, 113–115.

Rice-Evans, C. A., Miller, N. J., and Paganga, G. (1997). Antioxidant properties of phenolic compounds. *Trends in Plant Science*, 2(4), 152–159. https://doi.org/10.1016/S1360-1385(97)01018-2

Sasidharan, S., Chen, Y., Saravanan, D., Sundram, K. M., and Yoga Latha, L. (2011). Extraction, isolation and characterization of bioactive compounds from plants' extracts. *African Journal of Traditional, Complementary and Alternative Medicines*, 8(1), 1–10. https://doi.org/10.4314/ajtcam.v8i1.60483

Sinha, A. K. (1972). Colorimetric assay of catalase. *Analytical Biochemistry*, 47(2), 389–394. https://doi.org/10.1016/0003-2697 (72)90132-7

Yogananth, N., Buvaneshwari, S and Muthuezhilan, R. (2012). Larvicidal and antibacterial activities of different solvent extracts of *Solanum nigrum* Linn. *Global Journal of Biotechnology and Biochemistry* 7 (3): 86-89.

World Health Organization. (2000). *General guidelines for methodologies on research and evaluation of traditional medicine*. WHO.

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