Chapter 9

Phytochemistry, Antioxidant, Antibacterial Activity, and Medicinal Uses of Aromatic (Medicinal Plant *Rosmarinus officinalis*)

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Additional information is available at the end of the chapter

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Abstract

Rosemary is a well-known aromatic and medicinal plant whose consumption serves to remedy the number of disorders. Its essential oil (EO) constitutes an important ingredient for well-being feeling improvement through beauty products such as soaps, perfumes, and deodorants. The identification of phytochemical compounds is based on the peak area, retention time molecular weight, molecular formula, chemical structure, and pharmacological actions. It contains chemical constitutions, which may be useful for various herbal formulations as anti-inflammatory, analgesic, antipyretic, cardiac tonic, and anti-asthmatic. Therefore, this chapter reviews the phytochemical compounds of *Rosmarinus officinalis*, using methanolic extraction. The phytochemical compound is screened by gas chromatography-mass spectrometry (GC-MS) method and the evaluation of antimicrobial and antioxidant activities of the essential oils.

Keywords: aromatic and medicinal plant, *Rosmarinus officinalis*, gas chromatography-mass spectrum analysis, essential oil, antibacterial activity, antioxidant activity

1. Introduction

Rosmarinus officinalis thrives well in dry and arid regions, hills and low mountains, calcareous, shale, clay, and rocky substrates. Its use since ancient times in traditional medicine is justified by its antiseptic [1, 2], antirheumatic [3], anti-inflammatory, antispasmodic [4, 5], antimicrobial, and anti-hepatotoxic properties [6]. Its appreciation as a spice for seasoning and food preservation [7] is supported by a very high antioxidant activity [8]. The potent antioxidant properties of rosemary extracts have been attributed to its phenolic compounds, mainly rosmarinic acid and diterpenes carnosic acid and carnosol [9, 10]. Rosemary extract relaxes smooth muscles and has choleretic, hepatoprotective, and antitumorigenic activity [11]. Recent research shows that rosemary extracts possess strong anticancer properties. In the last few years, gas chromatography-mass spectrometry (GC-MS) has become firmly established as a key technological platform for metabolite profiling in plant [12–16]. GC-MS-based metabolome analysis has profound applications in discovering the mode of action of drugs or herbicides and helps unravel the effect of altered gene expression on metabolism and organism performance in biotechnological applications.

2. History

Rosemary has been named the Herb of the Year in 2001 by the International Herb Association. Hippocrates, Galen, and dioscorides prescribed rosemary for liver problems. Rosemary is not a popular plant in India. It was introduced by the Europeans as a garden plant due to its pleasant fragrant-scented leaves.

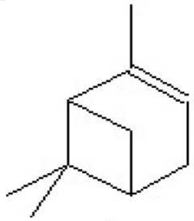
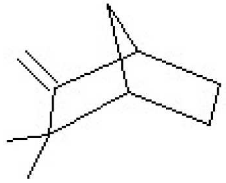
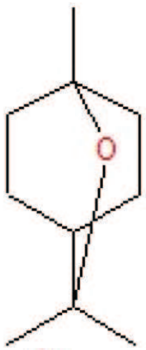
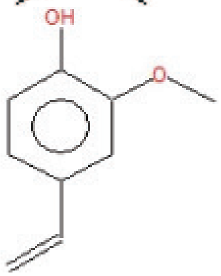
3. Varieties

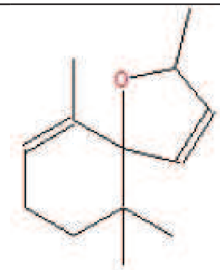
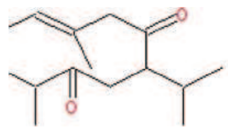
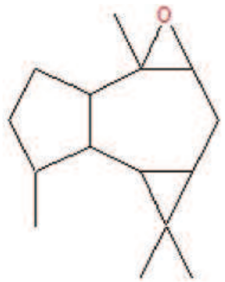
There are more than 20 varieties of rosemary plant. The different types of rosemary are as follows:

1. Upright rosemary: It measures between six and eight feet in diameter and two feet or more in height.
2. Creeping rosemary: It covers eight or 10 feet in diameter in a very short period of time. It can also trail down eight or 10 feet. It falls all the way to the ground and is covered with pale blue flowers.
3. Pine-scented rosemary: Pine-scented rosemary is a soft sea green that grows to about three to four feet high by about four or more feet wide.
4. Arp rosemary: This plant grows where winter temperatures are frequently in the teens or less.
5. Madalene hill rosemary: It is a cold hardy rosemary. It is rated to survive −15° and is erect, growing to about three feet. Its flowers are light blue.
6. Pink rosemary: It has the thinnest leaves of all *R. officinalis* plants. The flower is pale in color and grows quickly to two feet.
7. Dancing waters rosemary: It is shorter, more mounding and has dark blue flowers.
8. Golden rain rosemary: It has weeping foliage. The golden hue of the plant turns darker green over summer and returns with cooler weather.
9. Blue boy rosemary: It is the smallest of all the rosemary varieties. It has small leaves and little light blue pearls for flowers. This plant grows out to cover about 12 inches but rarely gets over six inches tall.

4. Microscopic characteristics

The leaf is dorsiventral with upper epidermal cells polygonal in shape [17].

Serial no.	Phytochemical compound	Formula	Exact mass	Chemical structure	Pharmacological actions
1.	α-pinene	$C_{10}H_{16}$	136.1252		Antimicrobial against bacterial and fungal cells activities
2.	Camphene	$C_{10}H_{16}$	136.1252		Antimicrobial against *Escherichia coli, Staphylococcus aureus*, and *Candida albicans*, but was not active against *Clostridium perfringens* up to the concentration of 100 g/ml. The significant antimicrobial and antioxidant activities of R. minima oil suggests that it could serve as a source for compounds with therapeutic potential
3.	Eucalyptol	$C_{10}H_{18}O$	154.13576		Eucalyptol, 1,8 cineole, is an essential oil present in large amounts in a variety of plants which is frequently used in the manufacture of cosmetics, to increase percutaneous penetration of drugs, as a nasal decongestant and anticough agent, in aromatherapy, and in dentistry (1-4). Eucalyptol has been used to treat bronchitis, sinusitis, and chronic rhinitis and also for the treatment of asthma
4.	2-Methoxy-4-vinylohenol	$C_9H_{10}O_2$	150.06808		Antioxidant and anti-inflammatory

Serial no.	Phytochemical compound	Formula	Exact mass	Chemical structure	Pharmacological actions
5.	1-Oxaspiro[4, 5] deca-3,6-diene,2,6,10,10-tetramethyl	$C_{13}H_{20}O$	192.151415		New chemical compound
6.	3-(*N*,*N*-Dimethyl lauryl ammonio) propanesulfate	$C_{17}H_{37}N_5O_3$	335.249414		New chemical compound
7.	Neocurdione	$C_{15}H_{24}O_2$	236.17763		Anti-viral, anti-bacteria and anti-tumor activity
8.	Isoaromadendrene epoxide	$C_{15}H_{24}O$	220.182715		Antibacterial activity and antioxidant activity

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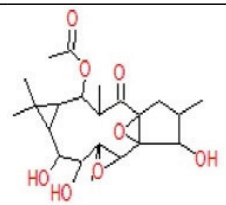
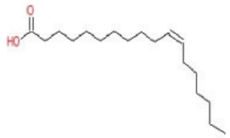
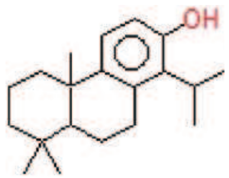
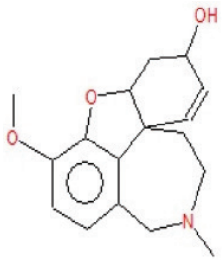
Serial no.	Phytochemical compound	Formula	Exact mass	Chemical structure	Pharmacological actions
9.	1b,4a-Epoxy-2H-cyclopenta[3, 4] cyclopropa[8, 9]cycloundec.	$C_{22}H_{32}O_8$	424.209419		New chemical compound
10.	cis-Vaccenic acid	$C_{18}H_{34}O_2$	282.25588		Anti-inflammatory
11.	2-Phenanthrenol,4b,5,6,7,8,8a,9,10-octahydro-4b,8,8-trimethyl-1	$C_{20}H_{30}O$	286.226999		Antimicrobial activity
12.	Galanthamine	$C_{17}H_{21}NO_3$	287.152143		Galantamine hydrobromide is a tertiary alkaloid drug that has been developed and approved in a number of countries including the USA and several countries in Europe as a treatment for mild-to-moderate Alzheimer's disease (AD)

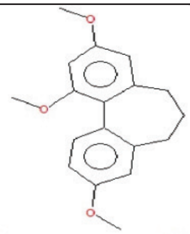
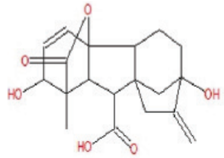
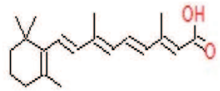
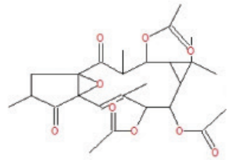
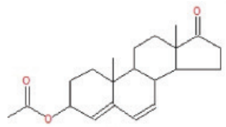
Serial no.	Phytochemical compound	Formula	Exact mass	Chemical structure	Pharmacological actions
13.	Dibenz[a,c] cyclohexane,2,4,7-trimethoxy	$C_{18}H_{20}O_3$	284.141245		New chemical compound
14.	2,4a,7-Trihydroxy-1-methyl-8-methyleneqibb-3-ene. 1,10-carboxylic acid.	$C_{19}H_{22}O_6$	346.141623		New chemical compound
15.	Retinoic acid	$C_{20}H_{22}O_2$	300.208931		Antibacterial activity and antioxidant activity
16.	7,8,12-Tri-O-acetyl-3-desoxy-ingol-3-one	$C_{26}H_{34}O_9$	490.220284		Antibacterial activity and antioxidant activity
17.	4,6-Androstadien-3β-ol-17-one,acetate	$C_{21}H_{28}O_3$	328.203844		Antioxidant activity and antibacterial activity

Table 1. Phytochemical compounds identified in methanolic extract of *Rosmarinus officinalis*

5. Major chemical constituents of *R. officinalis* using gas chromatography-mass spectrum analysis

The GC-MS analysis of the plant extract was made in a (QP 2010 Plus SHIMADZU) instrument under computer control at 70 eV [18–36].

Gas chromatography and mass spectroscopy analysis of compounds was carried out in methanolic seed extract of *R. officinalis*, shown in **Table 1**. Among the identified phytocompounds have the property of antioxidant and antimicrobial activities. Plant-based antimicrobials have enormous therapeutic potential as they can serve the purpose with lesser side effects [37–41]. In addition, rosemary harvested in Portugal is rich in myrcene (25%), 1,8-cineole, and camphor [42] while rosemary from North East of Spain presents an essential oil (EO) containing camphor and α-pinene as main constituents [43]. Furthermore, the essential oil of Lebanese rosemary is characterized by 1,8-cineole (20%) and α-pinene (18.8–38.5%) [44]. The major compounds of *R. officinalis*' essential oil from Eastern Cape Province in South Africa are verbenone (17.43%), camphor (16.57%), 1,8-cineole (11.91%), α-pinene (11.47%), borneol (5.74%), and camphene (5.70%) [45]. Many factors affect yield and chemical composition of essential oils such as drying, harvest period, harvest region, extraction technique, and the age of the plant [46, 47].

6. Antioxidant activity

Antioxidant activity of *R. officinalis* is due to its phenolic compounds including carnosic acid, carnosol, rosmarinic acid, and hydroxycinnamic acid ester. Rosemary uptake improves memory, and it is sometimes used as an antidepressant. It is also useful against cough and digestive disorders such as diarrhea, spasms, and flatulence. Thanks to diuretic and antispasmodics properties, the aerial parts of rosemary are orally used to relieve renal colic and dysmenorrhea [48–52].

7. Antibacterial activity

R. officinalis and *R. eriocalyx* EOs are extremely active on *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* strains. This result is very important especially for *P. aeruginosa*, which is known for its high resistance to all antibiotics. These results have some similarities with those of Taoufik Ouassil since he found that *R. officinalis* is active against the four species of bacteria (*Escherichia coli*: <14 mm, *Staphylococcus aureus*: <14 mm, *P. aeruginosa*: 14 mm, *K. pneumoniae*: 14 mm). Diameters of inhibition concerning *E. coli* and *S. aureus* are similar to ours but a great difference can be observed concerning *P. aeruginosa* and *K. pneumoniae*. Many researchers have highlighted sensitivity of Gram (+) bacteria compared to Gram (−) while testing natural extract but in our case it seems that Rosmarinus' essential oils are more active against Gram (−) bacteria. *R. officinalis* essential oil expressed a strong inhibitory activity against *K. pneumoniae* with an MIC of 2.08 mg/ml, and *S. aureus* with an MIC of 8.35 mg/ml. *E. coli* and

P. aeruginosa were inhibited with 16.7 mg/ml. *R. officinalis* EO has also a bactericidal power. Minimal bactericidal concentrations were 4.17 mg/ml for *K. pneumoniae* and 33.4 mg/ml for *E. coli, S. aureus*, and *P. aeruginosa*. According to our results, the MBC/MIC ratios are lower than four for all strains, so both essential oils have a bactericidal power against the tested strains. In Turkey (Izmir), Yesil Celiktas et al. (2007) worked on *R. officinalis* and found the following MIC: *E. coli* (20 mg/ml), *S. aureus* (10 mg/ml), *P. aeruginosa* (10 mg/ml), and *K. pneumoniae* (20 mg/ml). Okoh et al. [45] found that South African sample of *R. officinalis* (oriental region of the Cape) exhibited the following MIC: *E. coli* (7.5 mg/ml), *S. aureus* (3.75 mg/ml), and *K. pneumoniae* (0.94 mg/ml).

8. Brain, cardiovascular, gastrointestinal and other medicinal uses

It is used as carminative, rubifacient, and stimulant and as flavoring agent for liniments, hair lotions, inhaler, soaps, and cosmetics. Rosemary leaves have many traditional uses based on their antibacterial and spasmolytic actions. They are used orally for the treatment of dyspeptic complaints, and in external applications for supportive management of rheumatic complaints and circulatory disorders. *Aetheroleum Rosmarini* crude drug may enhance cognition. It is used as a cholagogue, diaphoretic, digestant, diuretic, emmenagogue, laxative, and tonic and also used in the management of headache, menstrual disorders, nervous menstrual complaints, tiredness, defective memory, sprains, and bruises:

1. Brain and nervous system conditions.
2. Cardiovascular conditions: It improves circulation, raises blood pressure, and stimulates the weak heart subject to palpitation when consumed in small doses.
3. Gastrointestinal circulatory systems: In conditions of bad breath, and stomach upset. Promotes proper digestion, toning, and calming effect on the digestion.
4. Reproductive system conditions: Stimulates the sexual organs.
5. Respiratory system: Colds and colic.
6. Other uses: The oil is used as perfume in ointments, shampoos, and soaps. The flowers are laid in clothes and cupboards to destroy moths. The leaves are crushed into meats, fish, potato salads, and so on.

9. Pharmacological properties

Singletary and Nelshoppen [53] studied the "Inhibition of 7, 12-dimethylbenz[c]anthracene (DMBA)-induced mammary tumorigenesis and of in vivo formation of mammary DMBA-DNA adducts by rosemary extract." Rosemary extract induces mammary tumorigenesis and in vivo formation of mammary dimethyl benz anthracene DNA adducts [54]. Hyperglycemic and insulin release inhibitory effects of *R. officinalis*. Krause et al. [55] studied the "Bioavailability of the antioxidative *R. officinalis* compound carnosic acid in eggs." Using this method, carnosic acid could be detected in 20 ng/g of egg yolk. Results showed that carnosic acid is bioavailable in egg yolk but not in albumen. Yen et al. [56] worked on the "Measurement of antioxidative activity in metal ion-induced lipid peroxidation systems." The antioxidant activity of α-tocopherol is less than that of rosemary extracts in the iron ion-induced peroxidation systems. Samman et al. [57] reported that "Green tea or rosemary extract added to foods reduces non-heme-iron absorption." The presence of the phenolic-rich extracts resulted in decreased non-heme-iron absorption [58]. Haloui et al. [59] studied the effects of aqueous extracts of the crude drug on the treatment of kidney function and diuresis in rats was determined. Jaswir et al. [60] studied "The synergistic effects of rosemary, sage, and citric acid on fatty acid retention of palm olein during deep-fat frying." A combination of 0.076% oleoresin rosemary extract, 0.066% sage extract, and 0.037% citric acid produced the optimal retention of the essential fatty acid [61]. Sotelo-Félix et al. [62] worked on the evaluation of the effectiveness of *R. officinalis* (Lamiaceae) in the alleviation of carbon tetrachloride-induced acute hepatotoxicity in the rat. Histological evaluation showed that *R. officinalis* partially prevented CCl4-induced inflammation, necrosis and vacuolation. Park et al. [63] reported the "Neuroprotective effect of *R. officinalis* extract on human dopaminergic cell line, SH-SY5Y. *R. officinalis* might potentially serve as an agent for the prevention of several human neurodegenerative diseases caused by oxidative stress and apoptosis. Sacchetti et al. [64] worked on the "Comparative evaluation of 11 essential oils of different origin as functional antioxidants, antiradicals and antimicrobials in foods." Antioxidant and radical-scavenging properties were tested by means of 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay, b-carotene bleaching test, and luminol-photochemiluminescence (PCL) assay. Cavero et al. [65] reported the "*In vitro* antioxidant analysis of supercritical fluid extracts from rosemary (*R. officinalis* L.)." Using forward stepwise multiple linear regression, carnosic acid, methyl carnosate, and carnosol were the compounds selected to predict the mentioned activity, with a value of 0.95 for the coefficient of determination. Antioxidant, antibacterial, and antifungal activities of the extracts were confirmed [66]. Moghtader et al. [67] reported "The evaluation of antioxidant potential of *Veronica officinalis and R. officinalis* extracts by monitoring malondialdehyde and glutathione levels in rats." The reduced and total glutathione were quantified from rat plasma, after derivatization with o-phthalaldehyde, using a high-performance liquid chromatography (HPLC) method with florescence detection. Salido et al. [68] studied the "Oxidative stress modulation by *R. officinalis* in CCl4-induced liver cirrhosis." The effect produced by a methanolic extract of *R. officinalis* on CCl4-induced liver cirrhosis in rats was investigated using both prevention and reversion models.

10. Conclusion

R. officinalis is the native plant of Iraq. It contains chemical constitutions which may be useful for various herbal formulations as anti-inflammatory, analgesic, antipyretic, cardiac tonic, and antiasthmatic. The phytochemical screening of the species has highlighted that both plants contain flavonoids, tannins, sterols and triterpenes, saponins, free anthraquinones, mucilages, cardiac glycosides, and catechols. Preliminary results of antibacterial study showed in vitro efficiency of *R. officinalis* and *R. eriocalyx* on all tested bacteria with minimum inhibitory concentrations ranging from 1.04 to 16.7 mg/ml. The results presented here may contribute to the knowledge of antimicrobial potential of these species. Other studies on extracts activities of these species are needed to compare them with essential oils activity. Rosemary is an exotic evergreen shrub with multiple medicinal and cosmetic properties. It is a popular herb which serves as flavoring agent and spice. Although it is well renowned for all these potencies, the oil of the plant is adhered with many side effects and hence lacks safety data. Therefore, the use of rosemary in pediatrics, as well as in pregnant women, should be always dealt with utmost care. It could be concluded that *R. officinalis* displays a wide variation in essential oil chemical composition in correlation with the climatic conditions under which it is grown, as well as the genetic variation, thus generating different chemotypes.

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