

Antibacterial Effects of Aqueous and Ethanolic Extracts of Medicinal Plants Against Pathogenic Strains

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Abstract

Background: Medicinal plants have been known since ancient times, as an anti-microbial spectrum against microorganisms. The production of secondary plant metabolites creates an important source of bactericide and various pharmaceutical drugs. In this study, the antimicrobial activity of aqueous and ethanolic extracts of the leaves of *Glycyrrhiza glabra*, *Mentha spicata* and *Rosmarinus officinalis* was evaluated against resistant strains of *Pseudomonas aeruginosa*, *Escherichia coli* and *Staphylococcus aureus*.

Methods: Antimicrobial effects of plants were investigated using a strain of *Pseudomonas aeruginosa*, *Escherichia coli* and *Staphylococcus aureus*, at the standard bacterium laboratory of the Veterinary Department, University of Islamic Azad, Tehran, Iran.

Results: Minimum inhibitory concentration (MIC) values ranged from 6.25 to 100.0 ppm, while Minimum bactericidal concentration (MBC) ranged from 12.5 to 200 ppm. The ethanol extract of *Glycyrrhiza glabra* had the lowest MIC (6.25 ppm), while the ethanol extract of *Rosmarinus officinalis* and *Mentha spicata* had the highest MIC (100 ppm), respectively. The MBC of the ethanolic extract of *G. glabra*, with the highest bacteria strains, was valuable against the lowest (12.5 PPM). The MBC of the ethanolic extract of *G. glabra* was valuable against the lowest bacteria strains.

Conclusions: Our findings showed good antibacterial activity for *G. glabra* extract that can be utilized as a treatment for infections, caused by *P. aeruginosa*, *E. coli* and *S. aureus*.

Keywords: *Glycyrrhiza glabra*, *Mentha spicata*, *Rosmarinus officinalis*, Antimicrobial Effect, Extract

1. Background

Medicinal plants have been known since ancient times, as an anti-microbial against a wide range of microorganisms. Nevertheless, medicinal plants were imported from the early 20th century (1-11). The production of secondary plant metabolites creates an important source of bactericidal and various pharmaceutical drugs (12, 13).

Rosmarinus officinalis belongs to the mint family. It is a perennial shrub with a pleasant-smell that grows in different regions around the world (14). It is a well-known medicinal and traditional plant, which is generally used in pharmaceutical products, as a digestive, tonic, astringent and diuretic, and practically for mosaic (15) and urinary diseases.

Peppermint oil is a species of mint, native to most parts of Europe, Asia, and naturalized in parts of North and West Africa, North and South America, as well as Oceania (16). Piperita essential oil destroys *Salmonella* (17) and is a deterrent on *Candida albicans* (18). In earlier research, essential oil showed antifungal activity against food poisoning pathogens with no evidence of mutagenicity in Ames test (19).

Licorice, which can be extracted from Licorice root, has a sweet flavor. Licorice root extract has several medical applications, which are also employed in traditional herbal medicine. Excessive consumption of licorice may lead to side effects, and clinical use in patients with hypokalemia, should be avoided, otherwise unexplained muscle weakness may occur (20).

Pseudomonas aeruginosa is a multidrug-resistant pathogen, recognized early for its ubiquity, and its mechanism of advanced inherently antibiotic resistance (21). Strains of *E. coli* cause serious food poisoning, and they are responsible for product recalls due to food contamination (22). *Staphylococcus aureus* is still one of the five most common causes of hospital-acquired infections, which is often due to postoperative wound infection (23).

2. Objectives

This study aimed to evaluate the antimicrobial activity of aqueous and ethanol extracts of the leaves of *Glycyrrhiza glabra*, *Mentha spicata* and *Rosmarinus officinalis* against resistant strains of *Pseudomonas aeruginosa*, *Escherichia coli* and *Staphylococcus aureus*.

3. Methods

3.1. Plant Material

Extracts of *G. glabra*, *M. spicata* and *R. officinalis* were prepared utilizing a rotary device. After collecting the plants, they were washed and chopped for microbial testing. The extract was then dried in the shade prior to preparation.

3.2. Aqueous Extract

To prepare aqueous extract, 50 g of dry powder from *G. glabra*, *M. spicata* and *R. officinalis* was dissolved in boiling water (250 mL) for 30 minutes; it was followed by boiling, filtering and freeze-drying (24).

3.3. Preparation of Plant Extracts

To prepare plant extracts, 10 g of dry powder of the plant was placed in half a liter flask containing 100 mL of ethanol with 96% water. The content of the flask was kept at room temperature for 24 hours and mixed by a shaker with a speed of 130 rpm, and subsequently drawing paper was used. The solvent extracts were separated by a rotary device and filtered, using a vacuum pump. The obtained extract was weighed, and then dissolved in Dimethyl Sulfoxide (DMSO) solvent, and later kept in a refrigerator at 4°C before use (24).

3.4. Determination of Extraction Yield (Percentage Yield)

Yield (percentage, w/w) of all dry extract was determined as: $\text{yield (\%)} = (W1/W2) \times 100$, where W1 is the weight of the extract after drying, by freezing in empty Hey pipes, and W2 is the weight of the powdered plant.

3.5. Isolates of Bacteria

Antimicrobial effects of the plant were investigated using a strain of *P. aeruginosa*, *E. coli* and *S. aureus*, at the standard bacterium laboratory of the veterinary department, university of Islamic Azad, Tehran, Iran.

3.6. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

Antibacterial activities of plant extracts were determined as described by Jahani (25).

Table 1. List of Medicinal Plants Utilized in This Study

Specie	Family	Organ Used
<i>Mentha spicata</i>	Lamiaceae	leaf
<i>Glycyrrhiza glabra</i>	Fabaceae	leaf
<i>Rosmarinus officinalis</i>	Lamiaceae	leaf

4. Results

This study tested antibacterial activity of medicinal plant extracts, belonging to three families of plants against pathogenic strains. These medicinal plants (Table 1) were selected based on two proposed anti-microbial activities, or previous studies, showing anti-bacterial properties using a variety of different extracts (15, 19, 20).

Table 2 shows water extract and ethanol plant. Extraction efficiency ranged from 20.1% to 41.6% and 22.8% to 38.8%. A wide range of yields, depending on the extraction solvent and the plant extract, was used. Peppermint oil extract exhibited the lowest yield of 18.1% (alcoholic extract). Maximum extraction efficiency was obtained from the leaves of *R. officinalis*, with 41.6% (aqueous extraction).

Table 2. Percentage of Extract Yield of Medicinal Plants Used

Plant Species	Extracta ^a	Yield (%)
<i>Rosmarinus officinalis</i>	A	41.6
	B	38.8
<i>Glycyrrhiza glabra</i>	A	22.8
	B	20.3
<i>Mentha spicata</i>	A	23.8
	B	20.1

^aExtract: A: aqueous; B: ethanolic.

Table 3 shows the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of active botanical extracts. The MIC and MBC of ethanol extract from each of the three species was established. Of all extracts, three showed bactericidal activity and three demonstrated antibacterial activities.

The MIC values ranged from 6.25 to 100.0 ppm while MBC values ranged from 12.5 to 200 ppm. The ethanol extract of *G. glabra* had the lowest MIC (6.25 ppm), while the ethanol extracts of *R. officinalis* and *M. spicata* had the highest MIC (100 ppm), respectively. The MBC of the ethanolic extract of *G. glabra* was valuable against the lowest bacterial strains (12.5 PPM, Table 3).

Table 3. Antibacterial Activity and Minimum Inhibitory Concentration (MIC) (Ppm) of the Extracts of *Glycyrrhiza glabra*, *Mentha spicata* and *Rosmarinus officinalis* Leaves^a

	<i>Mentha spicata</i>				<i>Glycyrrhiza glabra</i>				<i>Rosmarinus officinalis</i>			
	Aqueous extract		Ethanollic extract		Aqueous extract		Ethanollic extract		Aqueous extract		Ethanollic extract	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
<i>Pseudomonas aeruginosa</i>	-	-	25	50	-	-	25	50	-	-	25	50
<i>Escherichia coli</i>	-	-	25	50	-	-	6.25	12.5	-	-	50	100
<i>Staphylococcus aureus</i>	-	-	100	200	-	-	25	50	-	-	100	200

^a -: no activity.

5. Discussion

In this study, the acquisition of the aqueous and ethanol extracts was similar. However, greater efficiency in solution extraction is not related to higher antibacterial activity. Aqueous extracts showed no activity against pathogenic strains, as a result of incomplete extraction of active principles (26). Our findings are in agreement with Pinelo et al. (27), which suggested that the chemical properties of the solvent and method of extraction show distinct behavior.

Stomach ulcer properties, anti-inflammatory, immunomodulatory, anti-tumor and sputum licorice root were studied (28). Glycyrrhizin compound found in licorice, which is useful for liver protection, was suggested in treatment of tuberculosis; however, evidence for this use, which may be harmful, was not supported (29).

Glycyrrhizin also has anti-viral and anti-bacterial properties; the slow progress of Glycyrrhizin in intravenous viral hepatitis and autoimmune disorders (30) can also be supported. Licorice root in a clinical trial showed promising activity against atopic dermatitis (31). In addition, licorice root may be effective in the treatment of hyperlipidemia (32). Licorice root can also be effective in the treatment of inflammation-induced skin hyperpigmentation (33). Licorice root may also be useful in preventing neurological disorders (34).

5.1. Conclusion

Our findings showed good antibacterial activity of *G. glabra* extract that can be used as a treatment for infections, caused by *P. aeruginosa*, *E. coli* and *S. aureus*.

Footnotes

Authors' Contribution: All authors had an equal role in design, work, statistical analysis and manuscript writing.

Conflict of Interest: The authors declare no conflict of interest.

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