Research Article

In vitro evaluation of the antibacterial activity of ajwain oil on enteric pathogens

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ABSTRACT

Introduction: Essential oils are recently explored for many of their therapeutic potentials, and ajwail oil is traditionally well known for its diverse therapeutic uses. **Aim:** The aim of the present study was to evaluate the *in vitro* antibacterial activity of ajwain oil on selected enteric pathogens. **Materials and Method:** Different concentrations of ajwain oil (40, 80, and 100 µl/mL) were tested for its antibacterial activity against *Escherichia coli, Enterococcus Faecalis,* and *Klebsiella,* using agar well diffusion technique using Mueller-Hinton agar. The zone of inhibition was measured to find the antibacterial activity of ajwain oil. Amikacin was used as the positive control. All the tests were done in triplicate to minimize the test error. **Results:** The study showed that ajwain oil exhibits antibacterial activity against the enteric pathogens in a dose-dependent manner, and the maximum activity was shown against *E. coli* with a zone of inhibition of 40 mm at the maximum dose used for the study. **Conclusion:** The antibacterial activity of ajwain oil against these microorganisms provides a scientific background for its use in infections caused by the tested organisms. The presence of thymol in the ajwain oil may be responsible for its antibacterial effect against *E. coli, E. faecalis,* and *Klebsiella*.

KEY WORDS: Agar well diffusion, Ajwain oil, Enteric pathogens, *Trachyspermum ammi*

INTRODUCTION

Enteric infections are a major concern in the developing countries due to poor hygiene and sanitation.[1] To tackle this, epidemic improved sanitation and better health-care facilities are required, and thus, the search for new drugs is essential to reduce and control morbidity and mortality. The Indian tradition holds on to herbal medicine for centuries.[2] It has become the new alternative treatment option globally. People today do not rely on traditional beliefs that lack scientific backup.[3] Hence, traditional herbal medicine has gained popularity among the upcoming generation and medical professionals. Increased toxicity and side effects of allopathic medicine have driven toward more natural options that are safe, effective, and affordable forms of health care.[4,5] The curiosity and the search for truth have led to increased research in herbal medicines making it a part of everyday treatment.[6]

Medicinal plants continued to be excellent sources of phytochemicals and have versatile applications in traditional medicines, modern medicines, folk medicines, and food supplements.[7] Plant parts such as flower, buds, seed, leaves, and fruits are used for the isolation of essential oils and are mainly composed of monoterpenes, sesquiterpenes, and other isoprenes. As they have diverse bioactivity, they can be used for their antimicrobial, cytotoxic, and many other activities.[8-11]

Trachyspermum ammi L. (family *Apiaceae*) commonly known as ajwain is an annual herb found in India and Iran.[12] Ajawain seed is known for its antimicrobial, antiviral, nematicidal, antiulcer, antihypertensive antiseptic, stimulant, diuretic, and anesthetic properties.[13] Ajwain seed is rich in many phytochemicals, minerals, fiber, and carbohydrate. Ajwain fruit is reported to have thymol, para-cymene, γ-terpinene, α- and β-pinenes, dipentene, α-terpinene,

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Table 1: The zone of inhibition produced by different concentrations of ajwain oil on tested organisms

Concentration of ajwain oil (µL/ml)	Zone of inhibition (mm)		
	E. coli	*Klebsiella*	*E. faecalis*
40	28	22	21
80	32	29	28
100	40	37	35
Control	35	33	28

E. coli: Escherichia coli, E. faecalis: Enterococcus faecalis

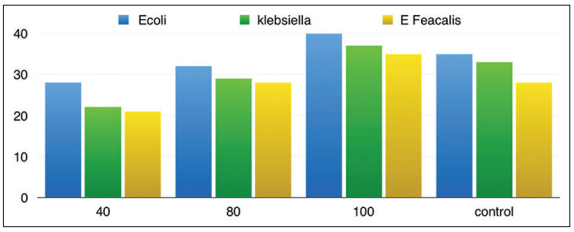

Figure 1: The antibacterial activity of ajwain oil at different concentrations

and carvacrol. The volatile oil contains thymol, γ-terpinene, para-cymene, and α- and β-pinenes.[14,15] Hence, in the present study, the antibacterial activity of the ajwain oil was studied against enteric pathogens.

MATERIALS AND METHODS

Plant Material

Ajwain oil from the seeds of *T. ammi* was used.

Test Organisms

The bacterial strains such as *Escherichia coli, Enterococcus faecalis,* and *Klebsiella* were used in the study. The organisms were obtained from the Department of Microbiology, Saveetha Dental College, and maintained in nutrient agar slope at 4°C.
Control antibiotic: Amikacin
Culture medium: Mueller-Hinton Agar.

Antibacterial Activity Assay

The bacterial suspension was standardized using the CLSI guidelines and was grown in Mueller-Hinton Broth for 18–24 h at 37°C followed by the matching of bacterial suspension to the turbidity equivalent to 0.5 McFarland solution. Evaluation of the antibacterial effect of ajwain was carried out by agar well diffusion assay. Different concentrations of the ajwain oil (40, 80, and 100 µg/mL) were used for the study. Lawn culture of the test organisms was made on the Mueller-Hinton agar (HiMedia) plates using sterile cotton swab, and the plates were dried for 15 min. Wells of 6-mm diameter was made using sterile cork borer, and 50 µl of different concentrations of the oil was filled in the well using micropipette on each of the plates containing cultures of the different bacterial strains. The culture plates were allowed to stand on the working bench for 30 min for pre-diffusion and were then incubated in an upright position at 37°C for 24 h. After 24 h, antibacterial activity was determined by measurement of the diameter of zones of inhibition.[1,16] Amikacin was used as control. All the tests were done in triplicate to minimize the manual error.

RESULTS

The antibacterial activity of the oil at different concentrations was screened against the microbes, and the zone of inhibition was measured in mm diameter. All the tested organisms such as *E. coli*, *Klebsiella,* and *E. faecalis* were inhibited by the ajwain oil in a dose-dependent manner with a maximum zone of inhibition of 40, 37, and 35 mm, respectively, at 100 µl/ml [Table 1 and Figure 1]. *E. coli* was most sensitive to the antibacterial effect of ajwain oil.

DISCUSSION

The presents study showed a concentration-dependent increase in the antimicrobial effect of the oil Hence, ajwain oil may be effectively used against infections caused by *E. coli, Klebsiella,* and *E. faecalis*. It was more effective against *E. coli*. Ajwain oil is also known for its antibacterial activity against foodborne pathogens.[17] Ajwain oil is reported to have thymol and carvacrol, which can impair membrane integrity and cause antibacterial activity.

There is an increase in drug-resistant pathogens due to an increased use of antibiotics in recent years. It is a major problem worldwide. Hence, the search for new antimicrobial agents has increased recently, and many plants are explored for their antimicrobial activity. Plants such as *Aesculus hippocastanum*, *Ficus racemosa, Glycyrrhiza glabra,* and *Solanum verbascifolium* Linn. are proven to have their antimicrobial activity and are traditionally used for many infections.[18-21] *Garcinia mangostana* Linn. and *Acacia catechu* Willd have effect against enteric pathogens.[1,22] Scientific exploration on more and more plants can bring new phytochemicals for the management of infections in a traditional way with minimal side effects and better patient compliance.

CONCLUSION

The present study revealed that ajwain oil has great potential against enteric organisms and may be used for the treatment of infections caused by these sensitive organisms. The presence of thymol and carvacrol in the ajwain oil may be the reason behind its antibacterial effect. Hence, this study can support the traditional use of ajwain for infections caused by enteric pathogens.

REFERENCES

1. Geetha RV, Roy A, Lakshmi T. Evaluation of antibacterial activity of fruit rind extract of *Garcinia mangostana* Linn. on enteric pathogens-an *in vitro* study. Asian J Pharm Clin Res 2011;4:115-8.
2. Vaidya AD, Devasagayam TP. Current status of herbal drugs in India: An overview. J Clin Biochem 2007;41:1-11.
3. Firenzuoli F, Gori L. Herbal medicine today: Clinical and research issues. Evid Based Complement Alternat Med 2007;4:37-40.
4. Verma S, Singh SP. Current and future states of herbal medicine. Vet World 2008;1:347-50.
5. Cheng YC. Why and how to globalize traditional Chinese medicine. J Tradit Chin Med 2011;1:1-4.
6. Dave PH, Vishnupriya V, Gayathri R. Herbal remedies for anxiety and depression-a review. Res J Pharm Tech 2016;9:1253-6.
7. Abdel-Hameed ES, Bazaid SA, Al Zahrani O, El-Halmouch Y, El-Sayed MM, El-Wakil AE. Chemical composition of volatile components, antimicrobial and anticancer activity of *n*-hexane extract and essential oil from *Trachyspermum ammi* L. seeds. Orient J Chem 2014;30:1653-62.
8. Adorjan B, Buchbauer G. Biological properties of essential oils: An updated review. Flavour Fragr J 2010;25:407-26.
9. Hamdy AE, Ahmed HE, Takayuki S. Bioactivity of essential oils and their volatile aroma components: Review. J Essent Oil Res 2012;24:203-12.
10. Bhardwaj P, Alok U, Khanna A. *In vitro* cytotoxicity of essential oils. Inter J Res Pharm Chem 2013;3:675-81.
11. Andrade B, Barbosa L, Probst I, Júnior A. Antimicrobial activity of essential oils. J Essent Oil Res 2014;26:34-40.
12. Wadikar D, Premavalli KS. Ajowan (*Trachyspermum ammi*) munch: A shelf stable ready-to-eatappetizer, its development and storage. Int Food Res J 2012;19:321-5.
13. Sahu S, Rawat D, Singh D. Antimicrobial activity of *Trachyspermum ammi* leaves mediated silver nanoparticles: Green approach. Int J Res Eng Technol 2015;4:35-8.
14. Bairwa R, Sodha RS, Rajawat BS. *Trachyspermum ammi*. Pharmacogn Rev 2012;6:56-60.
15. Nagalakshmi S, Shankaracharya NB, Naik JP, Rao LJ. Studies on chemical and technological aspects of ajowan (*Trachyspermum ammi* syn. Carum copticum). J Food Sci Technol 2000;37:277-81.
16. Jastaniah SD. The antimicrobial activity of some plant extracts, commonly used by Saudi people, against multidrug resistant bacteria. Life Sci J 2014;11:78-84.
17. Gandomi H, Abbaszadeh S, Jebellijavan A, Sharifzadeh A. Chemical constituents, antimicrobial and anti-oxidative effects of *Trachyspermum ammi* essential oil. J Food Process Preserv 2014;38:1690-5.
18. Roy A, Geetha RV, Lakshmi T. Evaluation of anti-mycotic activity of aqueous and ethanolic extracts of *Aesculus hippocastanum* -an *In vitro* Study. Int J Drug Dev Res 2011;3:335-8.
19. Bharathi S, Roy A. Antibacterial activity of the ethyl acetate extract of *Ficus racemosa* fruit on *Enterococcus faecalis*. Int J Pharm Sci Rev Res 2016;40:170-2.
20. Geetha RV, Roy A. *In-vitro* evaluation of the antibacterial activity of ethanolic root extract of *Glycyrrhiza glabra* on oral microbes. Int J Drug Dev Res 2012;4:161-5.
21. Roy A, Geetha RV. Evaluation of the antibacterial activity of the ethanolic extract of *Solanum verbascifolium* Linn -an *In vitro* Study. Res J Pharm Technol 2013;6:1359-13.
22. Lakshmi T, Geetha RV, Roy A. *In vitro* anti-bacterial activity of ethanolic bark extract of *Acacia catechu* wild against enteric pathogens. Int J Drug Dev Res 2011;3:328-34.

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