ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 11, Issue 2, 2018

Online - 2455-3891
Print - 0974-2441

Research Article

SCREENING OF ANTIBACTERIAL ACTIVITY OF FIVE DIFFERENT SPICES (AJWAIN, CORIANDER, CUMIN, FENNEL, AND FENUGREEK) AGAINST PATHOGENIC BACTERIAL STRAINS.

SALMA S*, LALITHA RAMAKRISHNAN, VINOTHINI J

Department of Nutrition and Dietetics, PSG College of Arts and Science, Affiliated to Bharathiar University, Coimbatore, Tamil Nadu, India. Email: smartsalma@outlook.com

Received: 18 August 2017, Revised and Accepted: 14 November 2017

ABSTRACT

Objective: This study was focussed on an evaluation of antibacterial activities of aqueous and alcoholic extracts of commonly consumed spices, namely, Ajwain (*Trachyspermum ammi*), Coriander (*Coriandrum sativum*), cumin (*Cuminum cyminum*), fennel (*Foeniculum vulgare*), and Fenugreek (*Trigonella foenum-graecum*).

Methods: This study includes the antibacterial effects of spices against six bacterial strains, namely, *Escherichia coli, Klebsiella pneumonia, Proteus vulgaris, Pseudomonas aeruginosa, Salmonella typhi,* and *Staphylococcus aureus* to compare their antibacterial effects by the paper disc agar diffusion method with three antibiotics such as amikacin, chloramphenicol, and vancomycin.

Results: According to findings, it is determined that inhibitory activity was detected on aqueous and alcoholic extracts of Ajwain, aqueous extract of cumin and on alcoholic mixed spice sample.

Conclusions: Among the five spices tested, only aqueous extracts of Ajwain and cumin exhibited antibacterial activity against one organism (*S. aureus*). Comparatively the alcoholic extracts gave a better response than the aqueous extracts. The effectiveness of the antibacterial activity was recorded better for the mixed spice samples when compared to that of the individual spices. This clearly emphasizes that the combined effect of the spices exhibited better antibacterial activity and the kill rate of the bacterial strains is higher relatively.

Keywords: Antibacterial activity, Ajwain, Cumin, Fennel, Fenugreek, Coriander, Bacterial strains.

 DOI: http://dx.doi.org/10.22159/ajpcr.2018.v11i2.22652

INTRODUCTION

Infectious diseases are the leading cause of morbidity and mortality worldwide, especially in developing countries. Infectious diseases are caused by microorganisms, such as bacteria, viruses, parasites, or fungi; the diseases that can be spread, directly or indirectly, from one person to another. The emerging of infectious diseases has been recognized as an important outcome of host-pathogen evolution leading to severe public health consequences [1].

Bacterial infections are a worldwide problem. In the past decade antibiotics, resistant infections occurred demanding new therapeutic strategies. For people living in developing countries, mainly medicinal plants and natural substances are available for the treatment of infectious diseases [2,3]. Foodborne pathogens such as *Escherichia coli* which are widely distributed in nature. It's implicated in large numbers of foodborne outbreaks in many parts of the world, including the developed countries [4]. The use and knowledge regarding traditional medicine are very diverse, and some traditional medicines were used by women to cure diseases and health care [5].

Early humans recognized their dependence on nature in both health and illness. Led by instinct, taste, and experience, and primitive men and women treated illness using plants [6]. Enormous advances have been made in medical care, but many people are still using herbal or alternative remedies [7].

Over the years and up to date, there have been numerous studies documenting the antibacterial, antifungal, antiviral, anticancer and anti-inflammatory properties of plant ingredients. Therefore, herbal derived substances remain the basis for a large proportion of commercial medications used today in developing countries [8]. This phenomenon has prompted researchers to identify alternative medicines, for the prevention and effective treatment of infections. Plant extracts and biologically active compounds isolated from plants have gained widespread interest, as they have the property to cure a variety of diseases [9].

Many of the spices and herbs used today have been valued for their antimicrobial effects and medicinal powers in addition to their flavor and fragrance qualities. Most of the foodborne bacterial pathogens examined were sensitive to spice extracts. The bacterial examination of spices and condiments are very important as they are used in the food preparation. These spices act through their natural inhibitory mechanisms and either inhibiting or killing the pathogens completely [10].

A spice is a dried seed, fruit, root, bark, or vegetative substance used in nutritionally insignificant quantities as a food additive for the purpose of flavoring, and indirectly for the purpose of killing and preventing the growth of pathogenic bacteria [11].

The organisms never remain the same, and they keep on change their characteristics. Some of the organisms do become resistant to antimicrobial components in the due course. Hence, a detailed study over the antimicrobial properties of spices would help us to know about the effectiveness of their inhibitory properties over certain organisms.

In this study, the antibacterial effects of five widely used spices in India such as Ajwain, coriander, cumin, fennel, and fenugreek were evaluated against the bacterial species such as *E. coli, Klebsiella pneumonia, Proteus*

Table 1: Comparison of antibacterial activity of spices

Name of the micro organism	Zone of inhibition (mm)											
	100 µl of aqueous extract						100 µl of alcoholic extract					
	A	B	C	D	E	F	A	B	C	D	E	F
E. coli	-	-	-	-	-	-	10	-	-	-	-	8
K. pneumoniae	-	-	-	-	-	-	11	-	-	-	-	6
S. typhi	-	-	-	-	-	-	9	-	-	-	-	-
S. aureus	8	-	9	-	-	-	-	-	-	-	-	7
P. vulgaris	-	-	-	-	-	-	13	-	-	-	-	6
P. aeruginosa	-	-	-	-	-	-	-	-	-	-	-	-

A: Ajwain, B: Coriander, C: Cumin, D: Fennel, E: Fenugreek, F: Mixed spice samples, *E. coli: Escherichia coli, K. pneumoniae: Klebsiella pneumoniae, S. typhi: Salmonella typhi, S. aureus: Staphylococcus aureus, P. vulgaris: Proteus vulgaris, P. aeruginosa: Pseudomonas aeruginosa*

vulgaris, Pseudomonas aeruginosa, Salmonella typhi, and *Staphylococcus aureus* and the inhibitory effects of these spices were compared with three antibiotics (amikacin, chloramphenicol, and vancomycin) and the results were discussed.

METHODS

Ethical standards

This article does not contain any studies with human participants or animals performed by any of the authors.

Microorganism

Standard strains of microorganisms used in the present study (*E. coli*, *K. pneumonia*, *P. vulgaris*, *P. aeruginosa*, *S. typhi,* and *S. aureus*) were obtained from the Department of Microbiology, PSG College of Arts and Science, Coimbatore.

Preparation of extracts

Good quality spices were purchased as samples from Nilgiris Departmental Stores at Coimbatore. To obtain the aqueous spice extract, about 5 g of each whole spice was powdered and added to 5 ml of distilled water. The solution is mixed well with a sterile glass rod. The extracts were sieved through a fine mesh cloth and then filtered. This extract was considered as the 100% concentration of the extract [12].

About 5 g of each spice was powdered as a whole at 25°C for 10 min and mixed with 70% ethanol. After centrifugation for 15 min, the supernatant solution was collected and utilized for the study [13].

For the preparation of mixed sample, 5 g each of all the five seed spices were taken instead of single seed spice. Then, the above mentioned aqueous and alcoholic extraction procedures were followed for the mixed spice sample extraction. The final filtrate obtained is known as the extract. The extracts were carefully transferred into the standard flasks and were stored in the cold room at 5°C.

Antimicrobial activity testing using dis diffusion method

The method of testing used for the present study is the disc diffusion method called as Kirby-Bauer method. The Kirby-Bauer method is based on the inhibition of bacterial growth measured under standard conditions. The organism to be tested is grown to a specific turbidity in a standard liquid medium. An inoculum from this culture is spread across the surface of a Mueller-Hinton agar plate to give confluent growth. Paper discs containing 100 µl concentration of each spice extract and standard antibiotics are placed on the agar surface.

The antibiotic in each disc diffuses outward from the disc, and the concentration of the antibiotic diminishes as the distance from the disc increases. After incubation at 35°C overnight, the diameter of the zone of growth inhibition is measured and scored according to the size of the zone. The size of the zone of inhibition is directly proportional to the sensitivity of the organism to the respective spice extract and the antibiotics.

Table 2: Comparison of antibacterial activity of standard antibiotics

Name of the micro organism	Antibiotics		
	Amikacin	Chloramphenicol	Vancomycin
E. coli	12	12	8
K. pneumonia	9	11	11
S. typhi	14	23	18
S. aureus	12	13	13
P. vulgaris	11	23	-
P. aeruginosa	15	18	8

E. coli: Escherichia coli, K. pneumonia: Klebsiella pneumonia, S. typhi: Salmonella typhi, S. aureus: Staphylococcus aureus, P. vulgaris: Proteus vulgaris, P. aeruginosa: Pseudomonas aeruginosa

According to Nagoba [14], the zones of inhibition of the antibiotics are compared with the zones of inhibition of test organisms and the results can also be interpreted based on the following criteria.

Sensitive

When zone diameter of test organism is greater than or equal to or not more than 4 mm, then the result can be interpreted as sensitive.

Medium sensitive

If the zone diameter is at least 12 mm but reduce in by more than 4 mm.

Resistant

If it shows no zone of inhibition of growth or if zone diameter is not >10 mm.

In this study, the zone of inhibition was observed for the aqueous extract of the spice samples and then the alcoholic extracts of the spice samples. Finally, the zones exhibited by the standard antibiotics (amikacin, chloramphenicol, and vancomycin) were measured, and the results were recorded. With the help of a simple ruler, the clear zone was measured. Care was taken not to measure the zone from the broken portion of the zone or the irregular lining of the zone. The clear zone which is completely transparent around the disc is the "zone of inhibition."

RESULTS AND DISCUSSION

This study was conducted to examine the inhibitory effects of spice extracts against some pathogens causing food poisoning and different illnesses in humans and some microorganisms causing spoilage in foods were used as test strains. For this purpose, the aqueous and alcoholic extracts of spices (Ajwain, coriander, cumin, fennel, and fenugreek) were tested on *E. coli*, *K. pneumonia*, *P. vulgaris*, *P. aeruginosa*, *S. typhi,* and *S. aureus* with disc diffusion method as *in vitro.*

Among the five spices tested, aqueous extracts of Ajwain and cumin exhibited antibacterial activity against one organism (*S. aureus*).

Alcoholic extract of Ajwain exhibited antibacterial activity against four organisms (*E. coli*, *K. pneumonia*, *P. vulgaris*, *P. aeruginosa*, and *S. typhi*). The alcoholic mixed spice sample exhibited antibacterial activity against four of the test organisms (*K. pneumonia*, *P. vulgaris*, *S. aureus,* and *E. coli*). Comparatively the alcoholic extracts gave a better response than the aqueous extracts.

The results of the antibacterial activity assays are represented in Tables 1 and 2.

On comparing the antibacterial activity exhibited by the standard antibiotics, the study reveals that the selected Ajwain sample has more antibacterial property and the next being the cumin sample whereas the mixed spice sample was at the best in exhibiting the antibacterial activity at the concentration of 100 µL. The other samples of coriander, fennel, and fenugreek lacked antibacterial property.

The effectiveness of the antibacterial activity was recorded better for the mixed spice samples when compared to that of the individual spices. This clearly emphasizes that the combined effect of the spices exhibited better antibacterial activity and the kill rate of the bacterial strains is higher relatively.

REFERENCES

1. Dhandapani K, Sonaimuthu V, Raghunath P. Investigation on the mechanism of action of the leaves of trianthema portulacastrum on human path only gens. Asian Journal of Pharmaceutical and Clinical Research 2016;9(3):135-140. Retrieved from: https://innovareacademics.in/journals/index.php/ajpcr/article/view/10690/5048.
2. Mulligan ME, Murry KA, Ribner BS, Standiford HC. Methicillin resistance *Staphylococcus aureus*. Am J Med 1993;94:313-28.
3. Al-Camo E. Chemotherapeutic Agents and Antibiotic Fundamental of Microbiology. 5th ed. U.S.A.: Adlison Wesley Longman Inc; 1996. p. 693-720.
4. Mead PS, Slutsker L, Dietz V, McCaig LF, Bresee JS, Shapiro C, *et al.* Food-related illness and death in the united states. Emerg Infect Dis 1999;5:607-25.
5. Lestari M, Fidrianny I, Yusuf A, Soemardji A. Brine shrimp teratogenic screening test on some selected plants. Asian Journal of Pharmaceutical and Clinical Research 2017;10(8):96-99. Doi: http://dx.doi.org/10.22159/ajpcr.2017.v10i8.18861.
6. Wink M. Biochemistry of plant secondary metabolism. Sheffield, U. K: Sheffield Academic Press; 1999. p. 205-13.
7. De Felippe Junior J. Plants com efeito no Diabetes mellitus. Diabet Med 2001;18:242-5.
8. Saiah H, Allem R, Kebir F. Antioxidant and antibacterial activities of six algerian medicinal plants. International Journal of Pharmacy and Pharmaceutical Sciences 2015;8(1):367-374. Retrieved from: https://innovareacademics.in/journals/index.php/ijpps/article/view/8220/3530.
9. Ponnuraj S, Jaganathan D, Kanagarajan M, Deivamarudachalam T. Spectroscopic analysis and antibacterial efficacy of bioactive compounds from limonia acidissima l. Fruit extract against clinical pathogens. International Journal of Pharmacy and Pharmaceutical Sciences 2015;7(3):383-389. Retrieved from. https://innovareacademics.in/journals/index.php/ijpps/article/view/4711/2084.
10. Sofia PK, Prasad R, Vijay VK, Srinivastava AK. Spices: Natural antimicrobials. J Spice India 2005;18:20.
11. Pelczar MJ, Reid R, Chan EC, Krieg NR. Microbiology. New York: McGraw-Hill Education Pvt. Ltd.; 1986.
12. Indu MN, Hatha AA, Abirosh C, Harsha U, Vivekanandan G. Antimicrobial activity of some of the south-Indian spices against serotypes of *Escherichia coli*, Salmonella, *Listeria* monocytogenes and *Aeromonas hydrophila*. Braz J Microbiol 2006;37:153-8.
13. Takikawa A, Abe K, Yamamoto M, Ishimaru S, Yasui M, Okubo Y, *et al.* Antimicrobial activity of nutmeg against *Escherichia coli* O157. J Biosci Bioeng 2002;94:315-20.
14. Nagoba BS, Pichare A. Medical Microbiology. St. Louis: Elsevier Publications; 2007. p. 117.