Original article

Evaluation of antibacterial activity of Indian spices against common foodborne pathogens

Papachan Karur Sofia,[1]* Rajendra Prasad,[1] Virendra Kumar Vijay[1] & Ashok Kumar Srivastava[2]

1 Center for Rural Development and Technology, Indian Institute of Technology, Hauz Khas, New Delhi, 110016, India
2 Department of Biochemical and Biotechnology, Indian Institute of Technology, Hauz Khas, New Delhi, 110016, India

(Received 6 July 2005; Accepted in revised form 19 April 2006)

Summary The present study was designed to evaluate the antimicrobial activity of six Indian spice extracts, namely clove, cinnamon, mustard, garlic, ginger and mint. All of these have been traditionally used in folk medicine, and are still used in the alternative system of health care. The antimicrobial activity of these commonly used Indian spices was tested against three potent foodborne pathogens, namely *Escherichia coli*,*Staphylococcus aureus* and *Bacillus cereus*, which are responsible for many health-related problems. These were tested using paper disc diffusion method, cup method and dilution method (qualitative). The results showed that the extracts of clove, cinnamon and mustard had good inhibitory action at 1% concentration, while garlic showed medium activity. At 3% concentration, complete bactericidal effect was achieved. Ginger and mint showed negligible antibacterial activity against these pathogens at the same concentration.

Keywords Antimicrobial activity, culinary, folk medicine, foodborne pathogens, Indian spices, medicinal plants, natural therapy, preservatives, sensory quality, zone of inhibition.

Introduction

India is one of the largest producer, consumer and exporter of spices. India grows over fifty spices out of the eighty-six grown worldwide. When one considers the need of its own vast population, India must be the world leaders *vis-à-vis* the area of cultivation of spices. Spices constitute an important group of agricultural commodity. It is virtually indispensable in the culinary art. They also play an important role in our national economy. On the world market, about twenty major spices are traded. They are included under raw crude medicinal plant materials in the export–import data (Pruthi, 1998).

For centuries, Indian spices have made a significant contribution both in the health care system and the food industry. Ancient Asian literature is a treasure of information related to the problems of health care and other environmental aspects. Indian spices have been used since ages in different traditional forms of medicine like Ayurveda, Unani and Sino Tibetian systems. The Vedic literature (2500 B.C.) is the main source of information that contributes to the development of Ayurveda. Particularly in Ayurveda, spices contributed a major amount for the treatment of key disorders of the body. Homeopathic medicine has been using spices as one of the chief ingredients in most of their preparations. In ancient India, natural herbs and spices were consumed either in food, or used as medicine in order to maintain proper sanitation, health and hygiene, and to increase longevity of life (De, 2004).

In this respect, spices, such as clove (toothache, fever, pain), cinnamon (nervous problems, stomach/intestine infections), mustard, garlic (antiseptic, diuretic), ginger (digestive aid, cold), mint etc. have been reported to possess very good medicinal properties. Apart from being a major part of the Indian culinary, spices also contribute to the modern allopathic system of healthcare by providing large number of medicines or parent compounds. Reports indicate that spices have dual type of action. Short-term effects include inflammation, pain, heat, redness and swelling. Long-term effects include anti-inflammation, analgesic, antimicrobial, antioxidant and antimutagenic actions (Kalia *et al*., 1977; Rusia & Srivastava, 1988; Ayoub, 1989; Schneider & Kubelka, 1989; Gangrade *et al*., 1990; Mahajan *et al*., 1991; Arora & Bhardwaj, 1997; Arora & Ohlan, 1997; Arora, 1998).

An interrelationship between the health-benefiting properties of spices and their use in food needs to be scientifically re-established. Contamination of food caused by unsanitary practices compromises the health

*Correspondent: Fax: +91 011 26526135;
e-mail: sofiapk@rediffmail.com

doi:10.1111/j.1365-2621.2006.01308.x

of people at various levels. Food safety, hence, becomes a key concern for the food-processing industry. Food safety enjoys a very low priority at all levels. There is limited awareness of its importance of hygiene and seriousness of foodborne illnesses. In addition to these hygiene practices, several food preparation techniques, such as preservation in spices have been used for thousands of years. The use of spices in traditional cooking is well known, and the antimicrobial effect of spices, such as turmeric is well documented (Marthi, 1999).

Studies on the control of pathogenic *Escherichia coli* and its species have been largely focussed on the use of chemical additives (Riley *et al*., 1983; Doyle, 1991; Zhao *et al*., 1993; Conner & Kotrola, 1995; Bari *et al*., 1999). Recent studies indicate that spices in low doses are beneficial to human beings (Banerjee & Sarkar, 2003a; De, 2004). Many food-associated outbreaks caused by *E. coli* have occurred from apple cider, beef, poultry, meat, milk etc. (Zottola & Smith, 1991). Pathogenic strains of *E. coli* cause hemorrhagic colitis, uraemic haemolytic syndrome and thrombotic thrombocytopenic purpurea (Zottola & Smith, 1991; Kim *et al*., 2004).

A large number of plants are used to combat different types of diseases and possess antimicrobial activity. In an era characterised by increasing consumer choice, self-medication and quest for natural therapy, herbal products are used increasingly as an alternative to drugs and supplements (Mansaray, 2000). In particular, extracts from many kinds of oriental spice plants are known to possess antimicrobial effect besides being used for the purpose of food preservation, appetiser promotion and medicinal purposes (Hoffman & Evans, 1911; Shelef *et al*., 1980; Zaika & Kissinger, 1981; Saleem & Ai Delaimy, 1982; Tassou *et al*., 2000; Yildrim *et al*., 2000). The essential oil of several plants shows activity against several bacteria, such as *Staphylococcus*, *Bacillus*, *Listeria* and *Kleisbella* (Baratta *et al*., 1998; Lafont *et al*., 1998). Arora & Kaur (1999) tested various spices for antimicrobial activity. Of the different spices tested, only garlic and clove were found to possess antimicrobial activity. Similarly, Conner & Beuchat (1984) studied essential oils of thirty-two spices for inhibitory effect on thirteen food spoilage and industrial yeasts. They found varying degree of inhibitory action amongst various spices; some being strongly antimicrobial, while others showing no antimicrobial activity at all.

Today, the exploration of naturally occurring antimicrobials for food preservation receives increasing attention. This is attributed to the consumer awareness of natural food products and a growing concern for microbial resistance towards conventional preservation (Gould, 1996). Hence this research is aimed at evaluating and validating the antimicrobial activity of Indian spices against common foodborne pathogenic bacteria.

Materials and methods

Experimental work

The antibacterial activity of six well-known and commonly used Indian spices, namely cinnamon, mustard, clove, ginger, mint and garlic was verified against three foodborne pathogens of significant importance, such as *E. coli*, *Staphylococcus aureus* and *Bacillus cereus*. All the selected spices were procured from the local market. They were categorised into dry and wet spices. Different parts of the plants were used enumerated as follows:

Name	Botanical name	Part of plant
Dry spices		
Clove	*Syzygium aromaticum*	Bud
Cinnamon	*Cinnamomum zeylanicum*	Bark
Mustard	*Brassica jancea*	Seed
Wet spices		
Garlic	*Allium sativum*	Bulb
Ginger	*Zingiber officinale*	Rhizome
Mint	*Mentha piperita*	Leaf

Preparation of plant/spice extracts

All the spices were first cleaned using tap water in order to remove any dirt or debris, and later using sterile distilled water. They were dried in laminar flow biological safety cabinet. Among the wet spices, garlic and ginger were skinned, while mint was destemmed. They were crushed in a sterile mortar pestle until a fine paste was obtained. Similarly, all the three dry spices were cleaned and a fine paste was made. Varying concentrations of the spice extracts (0.5, 1, 2 and 3% w/v) were made using sterile distilled water.

Preparation of bacterial cultures

Freeze-dried culture of three foodborne pathogens of *E. coli*, *S. aureus* and *B. cereus* were obtained from National Dairy Research Institute, Karnal, Haryana. These included NCDC 134, NCDC 133 and NCDC 250, respectively. The final concentration of the inoculum for the three micro-organisms used in the experiment was 10^8 CFU mL^{-1}, which was checked using the pour plate technique. For every experiment, freshly prepared sterile nutrient broth (10 mL) was inoculated from the slants.

Techniques

Paper disc diffusion technique (Kirby Bauer Technique)
Molten (45 °C) sterile nutrient agar (20 mL) was poured over base plates in sterile standard petri plates and

inoculated with 0.1 mL of each pathogenic strain by spread plate technique. The various concentrations of spice extracts were applied in 8-mm sterile filter paper discs (Whatman No. 1, 5 mm in diameter). The discs were placed on the surface of inoculated plates, and allowed to dry in a laminar flow biological safety cabinet. The sterile petri plates were then incubated for 24 h at 37 °C in an inverted position. The diameter of the zones of bacterial inhibition (with paper discs) was recorded. Control assay discs impregnated with sterile distilled water, without spices, were used. All the analyses were applied in triplicates (Black, 2002) (Table 1).

Cup/well method

The method given by Zaika (1988) was applied. Sterile nutrient agar (cooled to 45 °C) was poured into sterile standard petri plates (20 mL). This was then inoculated with 0.1 mL of pathogenic culture by spread plate technique. After setting, medium cups of 8 mm diameter were prepared with the help of a sterile cork borer. The base of each cup was sealed with 50 μL of sterilised molten nutrient agar. The cups were filled by adding 300 μL of the different spice extracts with varying concentration while sterilized distilled water was used as control. The plates were incubated for 24 h at 37 °C. After incubation, the zones of inhibition around each cup were measured (including cup) in millimetre with the help of antibiotic zone measuring scale. All the analyses were applied in triplicates (Table 2).

Dilution method for inhibitory effect and bactericidal effect (qualitative test)

Ten millilitre of each spice extract was inoculated with 0.1 mL of pathogenic culture and mixed well. Two-hundred microlitre of this mixture was pipetted into the micro array plates. After incubation at 37 °C for 24 h,

Table 1 Antibacterial activity of various concentrations of spices using paper disc diffusion technique

Sample	Concentration (%)	*Escherichia coli*	*Bacillus cereus*	*Staphylococcus aureus*
Garlic	Control	–	–	–
	0.5	10.0 ± 1.0	11.6 ± 0.5	10.6 ± 0.5
	1	10.0 ± 0.0	12.6 ± 0.5	12.0 ± 0.0
	2	11.3 ± 0.5	14.0 ± 0.0	12.3 ± 0.5
	3	12.3 ± 0.5	14.3 ± 0.5	14.0 ± 1.0
Ginger	Control	–	–	–
	0.5	9.3 ± 0.5	9.6 ± 0.5	10.6 ± 1.1
	1	10.3 ± 0.5	10.3 ± 0.5	11.3 ± 0.5
	2	10.6 ± 0.5	11.0 ± 0.0	13.0 ± 1.0
	3	11.3* ± 0.5	11.6 ± 0.5	14.6 ± 0.5
Mint	Control	–	–	–
	0.5	–	11.3 ± 0.5	12.6 ± 0.5
	1	13.3 ± 0.5	14.6 ± 1.1	14.3 ± 0.5
	2	15.0* ± 1.0	14.6* ± 0.5	15.0* ± 1.0
	3	16.3* ± 0.5	18.3* ± 1.1	17.3* ± 0.5
Clove	Control	–	–	–
	0.5	11.6 ± 0.5	10.3 ± 0.5	–
	1	13.3 ± 0.5	11.3 ± 0.5	–
	2	15.6 ± 0.5	12.6 ± 0.5	12.6 ± 0.5
	3	19.3 ± 0.5	16.0 ± 0.0	16.3 ± 0.5
Cinnamon	Control	–	–	–
	0.5	8.0 ± 0.0	10.3 ± 0.5	–
	1	10.0 ± 0.0	11.0 ± 0.0	–
	2	12.3 ± 0.5	11.3 ± 0.5	10.0 ± 0.0
	3	14.3 ± 0.5	12.0 ± 0.0	11.6 ± 0.5
Mustard	Control	–	–	–
	0.5	11.6 ± 0.5	–	10.3 ± 0.5
	1	19.3 ± 0.5	10.3 ± 0.5	11.6 ± 0.5
	2	21.6 ± 0.5	11.6 ± 0.5	14.3 ± 0.5
	3	25.6 ± 0.5	15.6 ± 0.5	16.0 ± 1.0

*Unclear zones of inhibition.
Differences between different concentrations within a spice found to be significant at $P < 0.05$.
All readings are mean of triplicates ± SD.

Table 2 Antibacterial activity of various spices: well or cup method

Sample	Concentration (%)	*Escherichia coli*	*Bacillus cereus*	*Staphylococcus aureus*
Garlic	Control	–	–	–
	0.5	10.3 ± 0.5	–	–
	1	12.3 ± 0.5	–	–
	2	13.0 ± 1.0	–	10.3 ± 0.5
	3	13.0 ± 0.0	10.6 ± 0.5	11.6 ± 0.5
Ginger	Control	–	–	–
	0.5	–	–	–
	1	–	–	–
	2	10.0 ± 0.0	10.3 ± 0.5	10.6 ± 0.5
	3	11.3 ± 0.5	11.3 ± 0.0	12.3 ± 0.5
Mint	Control	–	–	–
	0.5	–	13.3 ± 0.5	13.3 ± 0.5
	1	–	14.0 ± 0.0	14.6 ± 0.5
	2	8.6* ± 0.5	14.6* ± 0.5	26.6* ± 0.5
	3	9.3* ± 0.5	16.6* ± 0.5	27.6* ± 0.5
Clove	Control	–	–	–
	0.5	16.3 ± 0.5	–	–
	1	17.3 ± 0.5	14.6 ± 0.5	18.3 ± 0.5
	2	20.3 ± 0.5	23.6 ± 0.5	23.6 ± 0.5
	3	23.3 ± 0.5	25.6 ± 0.5	25.6 ± 0.5
Cinnamon	Control	–	–	–
	0.5	–	–	–
	1	10.6 ± 0.5	10 ± 0	10 ± 0
	2	12.6 ± 0.5	11.6 ± 0.5	10.6 ± 0.5
	3	14 ± 1	13.3 ± 0.5	12.3 ± 0.5
Mustard	Control	–	–	–
	0.5	8 ± 0	–	–
	1	12.6 ± 0.5	9.6 ± 0.5	–
	2	14.6 ± 0.5	10.3 ± 0.5	8.3 ± 0.5
	3	19.3 ± 0.5	11.3 ± 0.5	9.6 ± 0.5

*Unclear zones of inhibition.
Differences between different concentrations within a spice found to be significant at $P < 0.05$.
All readings are mean of triplicates ± SD.

the wells are checked for turbidity or spots/dots of bacteria or pellets formed at the base of the wells. Any growth at a particular concentration represented minimum inhibitory effect/minimum inhibitory concentration. Further, samples from the micro array plates that did not show any growth or turbidity were used to inoculate sterile fresh broth that contained no spice extracts. Therefore 100 μL of this culture was reintroduced into fresh (100 μL) nutrient broth. The lowest concentration of the spice extract that yielded no turbidity/spots or dots of growth following the second inoculation or sub-culturing showed the minimum bactericidal effect/minimum bactericidal concentration. For control plates, sterile distilled water was inoculated with bacterial strains without spices (Black, 2002).

Statistical analysis

Statistical analysis was carried out using ANOVA (two-factor) with replication. Mean and standard deviation were also calculated using the Microsoft Excel sheet, Office Edition 2000.

Results and discussion

In the present study, the antimicrobial activity of the six spice extracts, namely clove, cinnamon, mustard, garlic, ginger and mint were examined, both qualitatively and quantitatively by the presence or absence of bacterial growth, zone inhibition and turbidity.

Results from the antimicrobial disc diffusion and well method are summarised in Tables 1 and 2, respectively. Most of the spices showed good inhibitory activity against the three selected foodborne pathogens. In particular, mustard, clove and cinnamon showed strong antimicrobial activity at 1% concentration. Mustard was found to be most active against *E. coli*. It showed inhibition zones of up to 25.6 mm at 3% concentration. This antimicrobial activity in mustard can be attributed to allyl isothiocyanates – the principal active compounds of mustard. Rhee *et al.* (2003) also found mustard to have similar high inhibitory activity on *E. coli*, but in acidic products. Clove and cinnamon showed strong activity towards *E. coli* and *B. cereus*, but relatively less towards *S. aureus* at 0.5% and 1% concentrations. In a similar study, some researchers reported that cinnamon significantly reduced *E. coli* strains in apple juice (IFT, 1998). The potent antimicrobial activity of clove and cinnamon can be predominantly attributed to eugenol and cinnamaldehyde. These are the phenolic components of clove and cinnamon, which render them effective against the tested micro-organisms. This was confirmed by Farag *et al.* (1989), where eugenol, a major component of clove was found to limit the growth of *B. cereus* by inhibiting the production of certain enzymes needed for its growth. Two and three per cent clove concentrations showed inhibitory zones of up to 25.6 mm by the well method. Similar research on other bacteria studied by Bhak *et al.* (1990) showed cinnamon and clove to have strong inhibitory actions, while mustard and garlic had only slight antimicrobial activity. Table 1 clearly indicates that garlic was the most active against *B. cereus*. This could be contributed to Allium, one of the active principal components of freshly crushed garlic homogenates. It was also found to exhibit antimicrobial property against various species of *E. coli*. These results are corroborated by some researchers who believe that Allicin is the principal antimicrobial compound of freshly crushed garlic (Serge & David, 1999; Miron *et al.*, 2000). In a similar study carried out by Adler & Beuchat (2002), the addition of garlic to a food substrate enhanced the inactivation of *E. coli* at varying temperatures. In the present study, ginger showed its activity against *S. aureus*. However, as against most of the published studies, mint extracts did not show clear zones of inhibition. Differences within various spice concentrations were statistically significant at $P < 0.05$. The effectiveness of inhibitors can be sequenced as follows in descending order against different pathogens by disc diffusion assay:

Escherichia coli

Mustard > clove > cinnamon > garlic > ginger > mint

Bacillus cereus

Clove > mustard > cinnamon > garlic > ginger > mint

Staphylococcus aureus

Mustard > clove > cinnamon > ginger > garlic > mint

The effectiveness of inhibitors can be sequenced as follows in descending order against different pathogens by the well method:

Escherichia coli

Clove > mustard > cinnamon > garlic > ginger > mint

Bacillus cereus

Clove > cinnamon > mustard > ginger > garlic > mint

Staphylococcus aureus

Clove > cinnamon > mustard > ginger > garlic > mint

According to a review by Snyder (1997), similar observations were made where cloves, cinnamon and mustard were recognised as strong antimicrobial agents, while ginger and mint as weak ones. The inhibitory effect and bactericidal effect of the six selected spices were also evaluated with the help of micro array plates. As is evident from Table 3, among the wet spices only garlic showed a complete bactericidal effect (3%) against *E. coli*, whilst showing strong inhibitory effect (2% and

Table 3 Antibacterial activity of various spices using minimum inhibitory and bactericidal effect

Sample	Concentration (%)	*Escherichia coli*		*Bacillus cereus*		*Staphylococcus aureus*	
		MIC	MBC	MIC	MBC	MIC	MBC
Garlic	Control	–	–	–	–	–	–
	0.5	–	–	–	–	–	–
	1	++	–	–	–	–	–
	2	++	–	–	–	++	–
	3	++	++	++	–	++	–
Ginger	Control	–	–	–	–	–	–
	0.5	–	–	–	–	–	–
	1	–	–	–	–	–	–
	2	–	–	–	–	–	–
	3	–	–	–	–	–	–
Mint	Control	–	–	–	–	–	–
	0.5	–	–	–	–	–	–
	1	–	–	–	–	–	–
	2	++	–	++	–	–	–
	3	++	–	++	–	++	–
Clove	Control	–	–	–	–	–	–
	0.5	–	–	–	–	–	–
	1	++	–	++		++	++
	2	++	++	++	++	++	++
	3	++	++	++	++	++	++
Cinnamon	Control	–	–	–	–	–	–
	0.5	–	–	–	–	–	–
	1	++	–	–	–	–	–
	2	++	++	++	–	–	–
	3	++	++	++	–	++	–
Mustard	Control	–	–	–	–	–	–
	0.5	++	–	–	–	–	–
	1	++	–	–	–	–	–
	2	++	++	++	–	–	–
	3	++	++	++	–	++	–

–, no growth inhibition; ++, complete growth inhibition; MIC, minimum inhibitory concentration; MBC, minimum bactericidal concentration.

3%) against *B. cereus* and *S. aureus*. Our results are in accordance with Banerjee & Sarkar (2003b), who also found aqueous extracts of garlic to possess potent bacteriostatic principle against many bacteria at varying concentrations. These included *B. cereus* (6–10 mg mL^{-1}), *S. aureus* (30–40 mg mL^{-1}) and *E. coli* (30 mg mL^{-1}). Arora & Kaur (1999) also found that the antimicrobial effect of garlic extract was apparent within 1 h of incubation. The extract killed 93% of *Staphylococcus epidermis* and *Salmonella typhi* within 3 h. Ginger showed very mild inhibitory action against the three pathogenic bacteria, and was unable to show complete growth inhibition. Arora & Kaur (1999) confirm these results, where ginger showed little or no inhibition on different test bacteria. Mint was found to show better antimicrobial properties. It strongly inhibited (2% and 3%) the three foodborne pathogens. Among the dry spices, clove was the only spice to show complete bactericidal effect against all three foodborne pathogens at 3% concentration. Fabian *et al.* (1939) tested 10% extracts of cinnamon and clove against *B. subtilis* and *S. aureus*. They found cinnamon to be a slight inhibitor, while clove a strong inhibitor at 1:100 and 1:800, respectively. Aqueous clove infusions (0.1–1.0% w/v) and eugenol (0.06% w/v) inhibited overgrowth of germinated spores of *B. subtilis* in nutrient agar. Cinnamon and mustard were found to be bactericidal against *E. coli* only at 3%, while they only inhibited the other two bacteria at 3%. Similar results for mustard were seen against *Vibrio parahaemolyticus*, *Aspergillus* and *Penicillium* (Beuchat, 1976; Azzouz & Bullerman, 1982).

Conclusion

In conclusion, the results of the present study further demonstrated that clove, cinnamon, mustard, garlic, ginger and mint possess varying degree of antimicrobial activity. These spices act through their natural inhibitory mechanisms by either inhibiting or killing the pathogens completely. With the increasing awareness of people towards natural food and natural therapies, spices might act as the most obvious alternative. In developing countries like India, where spices are produced and used as food additives, their use as antimicrobial agents and potential preservatives can be extremely useful.

Acknowledgments

I would like to acknowledge the Center for Rural Development and Technology, IITD, where all the experimental work was carried out and the University Grants Commission for providing the research fellowship.

References

Adler, B.B. & Beuchat, L.R. (2002). Death of *Salmonella*, *E. coli o157:H7* and *Listeria monocytogenes* in garlic butter as affected by storage temperature. *Journal of Food Protection*, **65**, 1976–1980.

Arora, D.S. (1998). Antimicrobial activity of tea (*Cammelia sinensis*). *Antibiotic Chemotherapy*, **2**, 4–5.

Arora, D.S. & Bhardwaj, S.K. (1997). Antimicrobial activity of tea against some plant pathogens. *Geobios*, **24**, 127–131.

Arora, D.S. & Kaur, J. (1999). Antimicrobial activity of spices. *International Journal of Antimicrobial Agents*, **12**, 257–262.

Arora, D.S. & Ohlan, D. (1997). In vitro studies on antifungal activity of tea (*Cammelia sinensis*) and coffee (*Coffea arabica*) against wood rotting fungi. *Journal of Basic Microbiology*, **37**, 159–165.

Ayoub, S.M.H. (1989). Antimicrobial screening of Libyan medicinal plants. *Planta Medica*, **55**, 650–651.

Azzouz, M.A. & Bullerman, L.B. (1982). Comparative antimycotic effects of selected herbs, spices, plant components and commercial antifungal agents. *Journal of Food Protection*, **45**, 1298–1301.

Baratta, M.T, Dorman, H.J.D. & Deans, S.G. (1998). Chemical composition, antimicrobial and antioxidative activity of laurel, sage, rosemary, oregano and coriander essential oils. *Jornal of Essential Oil Research*, **10**, 618–627.

Banerjee, M. & Sarkar, K.P. (2003a). Microbiological quality of some retail spices in India. *Food Research International*, **36**, 469–474.

Banerjee, M. & Sarkar, K.P. (2003b). Inhibitory effect of garlic on bacterial pathogens from spices. *World Journal of Microbiology and Biotechnology*, **19**, 565–569.

Bari, M.L., Kusunoki, H., Furukawa, H., Ikeda, H., Isshiki, K. & Uemura, T. (1999). Inhibition of growth of *E. coli O157:H7* in fresh radish (*Raphanus sativus* L.) sprout production by calcinated calcium. *Journal of Food Protection*, **62**, 128–132.

Beuchat, L.R. (1976). Sensitivity of *Vibrio parahaemolyticus* to spices and organic acids. *Journal of Food Science*, **41**, 899–902.

Bhak, J, Yousef, A.E. & Martha, E.H. (1990). Behavior of *Listeria monocytogens* in the presence of clove oil. *Lebensm Wiss U Technol*, **23**, 66–69.

Black, J.G. (2002). *Microbiology Principles and Explorations*, 5th edn. Pp. 336–366. New York: John Wiley and Sons, Inc.

Conner, D.E. & Beuchat, L.R. (1984). Effects of essential oils from plants on growth of food spoilage yeasts. *Journal of Food Science*, **49**, 429–434.

Conner, D.E. & Kotrola, J.S. (1995). Growth and survival of *E. coli O157:H7* under acidic conditions. *Applied Environmental Microbiology*, **61**, 328–385.

De, A.K. (2004). *Spices: Traditional Uses and Medicinal Properties*. Pp. vii–xvii. Daryaganj: Asian Books Pvt Ltd.

Doyle, M.P. (1991). *E. coli O157:H7* and its significance in food. *International Journal of Food Microbiology*, **12**, 289–301.

Fabian, F.W., Krehl, C.F. & Little, N.W. (1939). The role of spices in pickled food spoilage. *Food Research*, **4**, 269–286.

Farag, R.S., Daw, Z.Y. & Abo-raya, S.H. (1989). Influence of some spice essential oils on *Aspergillus parasiticus* growth and production of aflatoxins in a synthetic medium. *Journal of Food Science*, **54**, 54–74.

Gangrade, S.K., Shrivastava, R.D., Sharma, O.P., Moghe, M.N. & Trivedi, K.C. (1990). Evaluation of some essential oils for antibacterial properties. *Indian Perfumer*, **34**, 204–208.

Gould, W.Ge. (1996). Methods of food preservation and extension of shelf life. *International Journal Food Microbiology*, **33**, 51–64.

Hoffman, C. & Evans, A.C. (1911). The use of spices as preservatives. *Journal of Indian Engineering Chemistry*, **3**, 835–838.

IFT (1998). *Spices may reduce E.coli O157: H7 in meat*. Institute of Food Technologist, http://www.sciencedaily.com/releases/1998/07980721081028.htm. Accessed on 12 May 2004.

Kalia, A.N., Chaudhary, N.C., Chugh, T.D. & Walia, S.K. (1977). Preliminary antimicrobial studies of Euphorbia. *Indian Drugs and Pharmaceutical Industries*, September–October, 1–3.

Kim, H.O., Park, S.W. & Park, H.D. (2004). Inactivation of *Escherichia coli O157:H7* by cinnamic aldehyde purified from Cinnamomum cassia shoot. *Food Microbiology*, **21**, 105–110.

Lafont, J., Jacyuet, J., Lafont, P., Romand, A. & Sarfasi, J. (1998). Some biological effects of spice, aromatics and condiments and other plant products on bacteria and micro mycelia. *Microbiologie-Aliments-Numtiox*, **2**, 239–249.

Mahajan, V., Arora, D.S. & Sabherwal, U. (1991). Antibacterial activity of some tea samples. *Indian Journal of Microbiology*, **31**, 443–445.

Mansaray, M. (2000). Herbal remedies-food or medicine? *Chemistry and Industry*, **20**, 677–678.

Marthi, B. (1999). Food safety challenges in developing countries: the Indian situation. *Food Control*, **10**, 243–245.

Miron, T., Rabinkov, A., Mirelman, D., Wilchek, M. & Weiner, L. (2000). The mode of action of allicin; its ready permeability through phospholipid membranes may contribute to its biological activity. *Biochemica et Biophysica Acta*, **1463**, 20–30.

Pruthi, J.S. (1998). *Spices and Condiments*, 5th edn. New Delhi: National Book Trust.

Rhee, M.S., Lee, S.Y., Richard, H., Dougherty, R.H. & Dong-Hyun, K. (2003). Antimicrobial effects of mustard flour and acetic acid against *E. coli O157:H7*, *Listeria monocytogenes* and Salmonella enterica. *Applied Environmental Microbiology*, **69**, 2959–2963.

Riley, L.W., Remis, R.S., Helgerson, S.D., Hargrett, N.T., Blake, P.A. & Cohen, M.L. (1983). Hemorrhagic colitis associated with a rare E.coli serotype. *The New England Journal of Medicine*, **308**, 681–685.

Rusia, K. & Srivastava, S.K. (1988). Antimicrobial activity of some Indian medicinal plants. *Indian Journal of Pharmaceutical Science*, **50**, 57–58.

Saleem, Z.M. & Ai Delaimy, K.S. (1982). Inhibition of *Bacillus cereus* by garlic Extract. *Journal of Food Protection*, **45**, 1007–1009.

Schneider, K. & Kubelka, W. (1989). The antibacterial activity of *Alchornea cordifolia*. *Planta Medica*, **55**, 651.

Serge, A. & David, M. (1999). Antimicrobial properties of allicin from garlic. *Microbes and Infection*, **2**, 125–129.

Shelef, L.A., Naglik, O.A. & Bogen, D.W. (1980). Sensitivity of some common food borne bacteria to the spice sage, rosemary and allspice. *Journal of Food Science*, **45**, 1042–1044.

Snyder, P. (1997). *Antimicrobial Activity of Spices and Herbs*. St. Paul, Minnesota: Hospitality Institute of Technology and Management, http://www.ift.org.

Tassou, C., Koutsoumanis, K. & Nychas, G.J.E. (2000). Inhibition of *Salmonella entridis* and *Staphylococcus aureus* in nutrient broth by mint essential oil. *Food Research International*, **33**, 273–280.

Yildrim, A., Mavi, A., Okty, M., Kara, A.A., Algur, O.F. & Bilaloggu, V. (2000). Comparison of antioxidant and antimicrobial activities of Tilia, Sage and Black tea extracts. *Journal of Agricultural Food Chemistry*, **48**, 5030–5034.

Zaika, L.L. (1988). Spices and herbs: their antimicrobial activity and its determination. *Journal of Food Science*, **9**, 97–118.

Zaika, L.L. & Kissinger, J.C. (1981). Inhibitory and stimulatory effects of oregano on *lactobacillus plantarum* and *Pediococcus Cerevisiae*. *Journal of Food Science*, **46**, 1205–1210.

Zhao, T., Doyle, M.P. & Besser, R.E. (1993). Fate of enterohemorrhagic *E. coli O157:H7* in apple cider with and without preservatives. *Applied Environmental Microbiology*, **59**, 2526–2530.

Zottola, E.A. & Smith, L.B. (1991). The microbiology of food borne disease outbreaks: an update. *Journal of Food Safety*, **11**, 13–29.