

ASSESSMENT OF ANTIBACTERIAL AND ANTIFUNGAL ACTIVITIES OF COMMON KITCHEN SPICES IN RAW AND PROCESSED FORMS

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Abstract

Spices have been known from centuries for its aroma, flavor and antimicrobial activities. This review illustrates antimicrobial activity of simple aqueous extracts of raw spices and commercially available processed spices against pathogenic strains of Staphylococcus aureus and Echerichia coli. Both raw and commercially available spices were extracted using distilled water and diluted at different concentrations of 0.2, 0.4, 0.6, 0.8 and 1.0 mg/ml. Antimicrobial activity of these extracts were tested using paper disc and agar cup diffusion method. Raw aqueous extract of garlic showed significant antimicrobial activity whereas dried and concentrated commercial garlic powder did not exhibit comparable antimicrobial activity. Other tested spices namely ginger and clove do not showed prominent zone of inhibition which fails to conclude a significant antimicrobial activity against the tested bacteria.

Keywords: Antimicrobial, Clove, Garlic, Ginger, Spices :

Introduction

Staphylococcus aureus is one of the most frequently identified pathogens in clinical laboratories. Infections caused by Staphylococcus aureus range from minor skin disorders such as wound infections, furuncles and carbuncles, and bullous impetigo, through locally invasive diseases such as cellulitis, osteomyelitis, sinusitis, and pneumonia, to major life-threatening septicemia and meningitis (Geo et al, 2010). This bacterium is also a frequent cause of medical device-related infections such as intravascular line sepsis and prosthetic joint infections. Although minor skin infections may resolve naturally without antibiotic intervention, once Staphylococcus aureus invades deeper structures, it often spreads hematogenously to other organ systems, leading to metastatic infection. Staphylococcal food poisoning occurs with a short incubation period of 2-6 h and is characterized by nausea and vomiting, that is followed by abdominal cramps and diarrhea, which can be hemorrhagic. It is mediated by enterotoxin B and occurs due to ingestion of food contaminated with preformed toxins (David et al, 2010)

Escherichia coli is a member of the normal flora of the human and animal gastrointestinal tract which helps in preventing the entry of pathogenic microorganisms. Under normal conditions, its presence is conducive to digestive processes. But when present in excess or in virulent form it causes diseases. Virulent strains of Escherichia coli can cause gastroenteritis, urinary tract infections, neonatal meningitis etc. (Mead et al., 1999). Traditional medicines have been used for many centuries by a substantial proportion of the

population of India. The natural products are found to be more effective with least side effects as compared to commercial antibiotics so that reason they are used as an alternated remedy for treatment of various infections. Natural products are a major source of new natural drugs and their use as an alternative medicine for treatment of various diseases has been increased in the last few decades (Vuorelaa et al., 2004; Ansari et al., 2006).

Spices are defined as plant substances used to enhance flavor. The active ingredients of plants against microorganisms are mostly some of the secondary metabolites (i.e. alkaloids, glycosides etc.) that are present in abundance in herbs and spices commonly used in Indian food preparations. (Tenover et al., 2004). Spices were used not only as flavouring agents, but also as food preservative. Certain spices prolong the shelf life of foods by preventing rancidity through their antioxidant property. Spices are rich in phytonutrients and other active ingredients that protect against disease and promote healing. In worldwide studies, spices have been linked to the prevention and treatment of chronic conditions such as heart disease, cancer, Type II diabetes, and

Alzheimer's. Unlike pharmaceutical drugs, spices can be used long term without concern for side effects (Beuchat, 1994).

With this background, the study was aimed to study the antimicrobial activity of simple aqueous extracts of raw and commercially available processed products against the pathogenic strains of *Staphylococcus aureus* and *Echerichia coli*.

Material and Methods

Bacterial culture

Staphylococcus aureus and *Echerichia coli* strains maintained at nutrient agar slant were used for the study. The culture was taken from the laboratory of Pillai college of Arts, Commerce and Science.

Maintenance of bacterial culture and inoculum preparation

Staphylococcus aureus and *Echerichia coli* strains maintained at nutrient agar slant were used for the study. Pure cultures were revived and maintained on nutrient agar slants and plates on regular basis. The cultures were streaked on sterile nutrient agar plates and kept in incubator for 24 hours at 37°C and stored at 4 °C. Bacterial cultures were sub-cultured after every 3 to 4 days to avoid contamination. Inoculum was prepared by growing the pure bacterial culture in nutrient broth over night at 37°C.

Preparation of aqueous extract for raw samples:

The spices were purchased from local market of Mumbai, India. Garlic and ginger were washed thoroughly with distilled water and 5g of each were homogenized using sterile mortar and pestle. And then sieved through double layer of sterile fine mesh cloth to make 100% extract. 5gm of dry clove were crushed and sieved through mesh cloth to get the fine powder. Powdered spices were soaked in 5ml of distilled water. All the tubes were centrifuged at 5000 rpm for 10 min. Filtrates were kept overnight at room temperature to extract the aqueous components. Different concentration of extract (0.2 mg/ml, 0.4mg/ml, 0.6 mg/ml, 0.8mg/ml and 1.0 mg/ml) from the above prepared stock of 1.0 mg/ml and are used for antimicrobial sensitivity assays.

For commercially available samples:

Packaged commercially available readymade spices powder and paste were also used. The whole Procedure remains the same as raw spices.

Antibacterial sensitivity testing**Disc diffusion method**

Filter paper disc of 5mm diameter using Whatman no. 1 filter paper was prepared and sterilized. The test microorganisms were transferred from nutrient broth to sterile Muller Hinton agar plates with the help of sterile cotton swabs. Filter paper disc were dipped in different concentration of spices extract and by using an ethanol dipped and flamed forceps the discs were aseptically placed over the Muller Hinton agar plates seeded with the test microorganisms.

Agar cup method

With the help of cork borer four cup were made per plate in the pre inoculated Nutrient-agar plates. Different extracts were added to these ditches (100 μ l) and plates were incubated at 37°C. After 24 hrs diameter of clear zone produced surrounding the ditches were measured to the nearest mm with the help of scale.

III. Results

Agar cup diffusion method was performed to study the antimicrobial activity of garlic, ginger and clove against *S. aureus* and *E.coli*. The relative size of the zone of inhibition determines the degree of sensitivity. Aqueous extract of garlic showed the significant antimicrobial activity while other tested spices do not give prominent zone of inhibition. Inhibitory effect of both the raw and commercially available aqueous extract of garlic was given below.

Table 1. Inhibitory effect of extracts of garlic on growth of *Staphylococcus aureus*.

Concentration (mg/ml)			Diameter Zone of Inhibition (mm)			Raw	Paste
0.2	14	-	0.4	22	-		
0.6	27	-	0.8	31	≤ 4		
1.0	37	≤ 7					

Table II. Inhibitory effect of extracts of garlic on growth of *Echerichia coli*

Concentration (mg/ml)		Zone of Inhibition (mm)	
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Raw Paste

0.2	12	-
		-
		-
		-
		≤5

0.4	17
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0.6	24
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0.8	28
-----	----

1.0	33
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- Indicates no zone of inhibition

The results indicated that the antimicrobial activity of raw garlic is stronger than the commercially available paste of garlic. The diameter of zone of inhibition increases with the increase in concentration of aqueous extract. Moreover, *Staphylococcus aureus* is found to be more sensitive than *Echerichia coli*. Paper disc diffusion method also give comparable data to that of agar cup method but the zone of inhibition was more prominent and of bigger size in agar cup method. Thus, extracts of garlic displayed excellent inhibition on the growth of gram positive and gram negative bacteria.

Discussions

Traditionally, spices are part of routine Indian food preparations as they make food appealing by providing better appearance, smell and taste. 'Ayurveda' the indigenous system of Indian medicine, uses a large number of spices in combinations as preventive and curative medicines (Ali et al., 2008). There are number of reports available in literature stating the sensitivity of various bacteria towards these herbs and spices used in food preparations (McNamara et al., 2005). Most of these studies involve extraction of the active component in the herbs and spices using organic solvents. But as an ingredient of food, extraction using distilled water is more reliable than organic solvent and it mimics the way we used to add the spices during cooking.

Therefore, in the present study the commonly used spices extracts are made with distilled water and tested for its antimicrobial effect against *Staphylococcus aureus* and *Echerichia coli*. Prominent zone of inhibitions seen with aqueous extract of raw garlic established a significant antimicrobial activity whereas dried and concentrated commercial garlic powder did not exhibit comparable antimicrobial activity. Other tested spices namely ginger and clove do not showed prominent zone of inhibition which fails to conclude a significant anti-microbial activity against the tested bacteria. Considering the above results, it can be suggested that addition of raw spices is more useful than the commercially available processed powder or paste to the food preparations which can help to keep a check on the concentration *Staphylococcus aureus* and *Escherichia coli* in the body.

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